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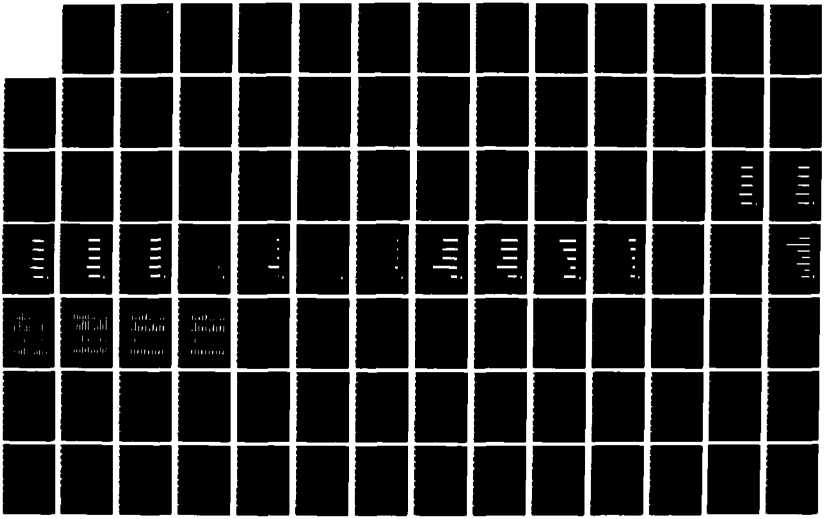

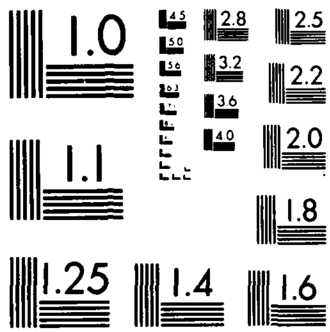

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Technical Report E06549-38
Contract No. N00039-84-C-0070

IITRI

COMPILATION OF 1986 ANNUAL REPORTS OF THE NAVY ELF COMMUNICATIONS SYSTEM ECOLOGICAL MONITORING PROGRAM

Volume 2 of 3 Volumes: TABS D-G

AD-A184 562

July 1987

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19. ABSTRACT (Continue on reverse if necessary and identify by block number)

This is the fifth compilation of annual reports for the Navy's ELF Communications System Ecological Monitoring Program. The reports document the progress of ten studies performed during 1986 at the Wisconsin and Michigan Transmitting Facilities. The purpose of the monitoring is to determine whether electromagnetic fields produced by the ELF Communications System will affect resident biota or their ecological relationships.

See reverse for report titles and authors.

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19. Abstract (Continued)

D. Soil Amoeba
Michigan State University
Band, R.N.

E. Soil and Litter Arthropoda and Earthworm Studies
Michigan State University
Snider, R.J.; Snider, R.M.

F. Biological Studies on Pollinating Insects: Megachilid Bees
Michigan State University
Strickler, K.; Scriber, J.M.

G. Small Vertebrates: Small Mammals and Nesting Birds
Michigan State University
Beaver, D.L.; Asher, J.H.; Hill, R.W.

FOREWORD

The U.S. Navy is conducting a long-term program to monitor for possible effects from the operation of its Extremely Low Frequency (ELF) Communications System to resident biota and their ecological relationships. The program is being implemented by IIT Research Institute (IITRI) under contract to the Space and Naval Warfare Systems Command (SPAWAR). IITRI provides engineering support and coordinates the efforts of investigators. Monitoring projects are being carried out through subcontract arrangements between IITRI and study teams at several universities.

This is the fifth compilation of annual reports prepared by university study teams. Each report chronicles the data collection and analysis activities for a monitoring project during 1986. As in the past, each report has been reviewed by four or more scientific peers. Investigators have considered and addressed reviewer critiques prior to providing their report for printing. Reports have been printed from original copies without change or editing by either IITRI or SPAWAR.

The 1986 compilation is one of a series that documents the activities of the Ecological Monitoring Program since its inception in 1982. Other reports document engineering support and summarize the progress of the Program. Previous reports provide information on the background, overall design, and early development of the Program. All of these reports have been provided to the National Technical Information Service for unlimited distribution. The results of monitoring activities have also been presented at scientific meetings or as journal articles.

Cont'd

ELF ECOLOGICAL MONITORING PROGRAM
INDEX OF 1986 ANNUAL REPORTS

This report documents progress of the following studies:

A. Herbaceous Plant Cover and Tree Studies
Michigan Technological University
Becker, C.; Brooks, R.; Bruhn, J.; Cattelino, P.; Connaughton, P.; Fuller, L.; Gale, M.; Holmes, M.; Jurgensen, M.; Lederle, K.; Liechty, H.; Mroz, G.; Reed, D.; Reed, E.J.; Richter, D.

B. Litter Decomposition and Microflora
Michigan Technological University
Bagley, S.; Bruhn, J.; Pickens, J.B.

C. The Effects of Exposing the Slime Mold <u>Physarum polycephalum</u> to Electromagnetic Fields
University of Wisconsin-Parkside
Goodman, E.M.; Greenebaum, B.

D. Soil Amoeba;
Michigan State University
Band, R.N.

E. Soil and Litter Arthropoda and Earthworm Studies;
Michigan State University
Snider, R.J.; Snider, R.M.

F. Biological Studies on Pollinating Insects: Megachilid Bees; and
Michigan State University
Strickler, K.; Scriber, J.M.

G. Small Vertebrates: Small Mammals and Nesting Birds.
Michigan State University
Beaver, D.L.; Asher, J.H.; Hill, R.W.

H. Aquatic Ecosystems
Michigan State University
Burton, T.M.; Stout, R.J.; Taylor, W.W.; Muzzall, P.M.; Oemke, M.P.; Glosser, R.; O'Malley, M.; Whelan, G.

I. Wetland Studies
University of Wisconsin-Milwaukee
Stearns, F.; Guntenspergen, G.; Keough, J.; Wikum, D.

J. Bird Species and Communities
University of Minnesota-Duluth
Hanowski, J.M.; Niemi, G.J.; Blake, J.G.

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East Lansing, Michigan 48824

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Department of Zoology

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Rudolph Neal Band, PI

f. Co-investigators: none

g. Name and signature of subcontractor's approving and releasing authority:

Howard G. Grider

Howard G. Grider, Director

Contract and Grant Admin.

3. Table of Contents:

4. Abstract:

The most outstanding aspect of the 1986 field season was the drought that extended from May through August, which was catastrophic to farmers in the U.P. It also suppressed population growth of soil amoebae.

Antenna and control sites, used in previous seasons were continued. A new ground wire site was used in 1986. The sites have been characterized as to electromagnetic background (by IITRI personnel), physical and chemical properties, and biological characteristics.

Studies of soil amoebae in 1983 were designed to provide sufficient data to determine sample sizes and methods of statistical analysis suitable for comparing control and experimental sites. These were utilized in the 1984, 1985 and 1986 field seasons. Fluctuations in total amoeba number were observed in both the 1984 and 1985 growing seasons at control, antenna and ground study sites. The 1986 drought suppressed significant growth of soil amoebae.

The genetic diversity within a species of soil amoeba was determined by isoenzyme analysis of clones isolated from control, antenna and ground sites. To test the effect of ELF electromagnetic radiation on the growth of amoebae culture chambers were designed and constructed by IITRI personnel. These were used at the Wisconsin Transmitter Facility in 1986.

5. Summary:

Plot selection and characterization: Site selection has been completed. Soil chemistry was performed on all sites in 1986, with a statistical comparison of sites. Unlike 1985, there were some differences between sites in terms of soil chemistry. Possibly this was due to the atypical, dry season.

Species and strain characterization: Acanthamoeba polyphaga was used to test for strain heterogeneity within the sites. Isoenzyme analysis was chosen as the technique to detect strain differences. Sufficient heterogeneity was observed between clonal isolates to make this a useful technique to detect possible effects by ELF electromagnetic radiation on heterogeneity within a species. A paper on this is in press in the J. Protozoology. Restriction fragment analysis of mitochondrial DNA is still being developed. We have submitted a paper on the taxonomy of Naegleria species using this technique.

Population size: The total population of amoebae increased during the 1984 and 1985 growing seasons. Toward the end of both growing seasons, the total population of amoebae decreased. Population sizes were the same at control, antenna and ground sites. The ratio between vegetative amoebae and dormant cysts was labile both within and between sites. The biological basis for population fluctuations are being investigated. Probably due to the drought, little growth was observed in the 1986 season although population size did not differ statistically between study sites. Therefore, even though 1986 was an unusual year, populations did not differ between study sites.

Growth and feeding activity: In the 1984 season, studies on growth in culture chambers by A. polyphaga demonstrated no significant difference in growth between sites. This was done to test the experimental and statistical means for examining growth "under the wire" and to test the culture chamber design. By the 1985 season, the electrical circuits associated with the culture chamber was designed and constructed by IITRI personnel. An attempt was made to test the unit at the Wisconsin Transmitter Facility, but it was not satisfactory because the antenna was not running long enough to do the experiments at those times chosen to work. In 1986 it was possible to do a growth experiment while the transmitter was running. The brief experiment failed to reveal differences in growth between control and experimental culture chambers, but it did provide an opportunity to test the system.

Ambient monitoring: Soil temperature and moisture were monitored continuously during the 1984 and 1985 field seasons (June through September). In both seasons the ambient data did not correlate to changes in amoeba population. The moisture content of soil in 1986 was lower and this correlated to small populations of amoebae in soil. Soil temperature was somewhat higher during the growing season, probably due to the drought.

6. Progress report:

OBJECTIVES: The project objective is to determine effects of ELF radiation on amoebae in soil. The sites chosen for this study are adjacent to the Michigan ELF transmitter.

For the 1986 field season, as was true for the 1984 and 1985 seasons, the primary objective was to demonstrate that the control, antenna and ground wire study sites were biologically similar in regards to soil amoebae. In addition a base line was accumulated for comparison with future data, especially that obtained once the antenna is operational.

WORK PLAN ELEMENTS:

#0. Plot selection and characterization.

Synopsis: The ground wire site was moved to conform with changes in the antenna ground location. From the data given in this report, it would appear that the new site is biologically similar to the other sites. Hopefully, site selection is now complete. Statistical analysis of soil chemistry shows variability between sites. This may have been due to the exceptionally dry season (Fig. 14); 1987 data may be more meaningful.

#1. Species and strain characterization.

Synopsis: using morphological and physiological markers, identify species and strains of soil amoebae from the study areas so that possible changes in the population due to ELF can be detected.

Specifics: Species of soil amoebae present at the study sites are isolated from soil enrichment plates. In this way, clonal isolates of A. polyphaga were obtained from control, antenna and ground sites for isoenzyme analysis. Soil amoebae are asexual organisms, reproducing without apparent sexual, genetic recombination. However, isoenzyme analysis reveals significant heterogeneity between clonal isolates.

The isoenzyme patterns are the same as those observed for sexually reproducing, diploid organisms. For this reason, the analytical and statistical techniques developed for isoenzyme analyses of higher organisms is used in the present study. There is a precedence for this approach, it has been used recently to examine laboratory isolates of Naegleria species (i.e. Pernin et al. 1985). The "genetic distance" between clonal isolates and between sites were determined by Nei's method (Nei, 1972), a widely used mathematical expression of the relationships between related organisms. This approach has been used to study inter and intra-species relationships (e.g. Avise et al., 1975). We have published a paper on the genetic heterogeneity observed at the study sites (Jacobson & Band, 1987).

It was suggested last year that I use species diversity/richness indices. This is not feasible, as noted in the 1984 annual report. Not enough personnel are available to do the work necessary for this analysis. Since all of the species observed do not appear on similar dilution plates (see the next work plan element), many more plates would have to be set-up than is required for counting amoebae. As pointed out by one of the reviewers, possible changes in species diversity would be preceded by changes in genetic heterogeneity within the species.

Mitochondrial DNA techniques are in progress. We have submitted a paper on mtDNA and evolutionary relationships within the genus *Naegleria*. A mtDNA analysis of *A. polyphaga* clonal isolates may be needed to demonstrate species relationships.

#2. Population size and activity.

Synopsis: determine population size of amoebae in soil and the ratio of vegetative to dormant amoebae over the growing season. This is a productivity measure which could be affected by ELF radiation, it could also be a reflection of changes in the microbial food organisms due to ELF radiation.

Specifics: an established soil dilution counting technique is used (Singh, 1946 as modified by Darbyshire *et al.*, 1974). In order to count vegetative amoebae and cysts, samples are first divided in half, one-half is used to count total cysts and vegetative amoebae while the other half is treated to kill amoebae so that only cysts are counted. Differential counts are used to calculate by subtraction the total vegetative amoeba count. In the 1983 season I found that 8 random samples, subdivided into organic and mineral horizons (i.e. 8 samples per horizon), provided statistically significant data. Two-way analysis of variance was used to demonstrate that there was no significant difference in total amoeba and cyst count between control, antenna and ground sites for each horizon in 1986. Table 4B gives the error (i.e. among) degrees of freedom as 21.

#3. Growth and feeding activity.

Synopsis. Determine the in situ growth and feeding activity of amoebae in soil submersible culture vessels. This will provide data on growth rate, feeding activity and mean generation time (i.e. the cell cycle between nuclear mitoses).

Rationale. The approach utilizes a known amoeba species previously isolated from the study site, Acanthamoeba polyphaga and characterized as part of the isoenzyme study. Direct counts of amoebae are made with a microscope to determine increase in number of organisms and nuclei over time. A log transform of these data provides a straight line plot which can be quantified by regression analysis. Statistically significant differences between slopes can be detected with confidence limits of the line, a version of the t-test. This approach will be used to determine growth rate and thus mean generation time. Mean generation time is comparable to the cell cycle measurement of time between mitoses of Physarum. Cropping activity will be determined by varying the number of bacteria for amoeba growth and then following growth rate and maximum yield. Culture chambers, containing electrodes to eventually use in conjuction with ELF induced currents, was designed with the help of IITRI personnel. These electrodes will be connected with electrodes buried in soil adjacent to the culture chambers, to produce the necessary voltage drop from the current in the soil, induced by ELF radiation.

In the 1984 field season, it was demonstrated that counts from chambers buried at the research sites yielded growth rates that were statistically the same. For the 1985 season, IITRI personnel designed and constructed the electrical components to interface between the soil electrodes and the culture chambers. See appendix for test setup provided by IITRI. It was not possible to test these under the Wisconsin Transmitter antenna in 1985 because it was only periodically operational. We were able to perform growth experiments in 1986 at the Wisconsin transmitter. The electrical interface between the culture chamber, containing electrodes to pass a current through the culture saline from soil electrodes, is necessary because it is not possible to mimic electrical properties of soil in a physiological saline. The soil electrodes consist of copper pipe, 4" diameter and 3' long. Soil water is a dilute saline, suitable for amoeba growth, but it is not a continuous phase across a significant space. Therefore soil exhibits higher resistance than would be the case for soil water. In the case of physiological saline in the culture chambers, the resistance across the electrodes is much lower than a comparable distance in soil. If growth in soil were used, it would mimic normal soil properties but cell counting procedures would lose a significant degree of accuracy since enrichment counting procedures would be required. Therefore, two different culture chamber arrangements are needed, one to mimic the voltage induced in soil and the other to mimic the current.

#4. Ambient monitoring.

Synopsis. Soil temperature and moisture are monitored. Both measures are useful for general trends but failed to correlate to changes in amoeba populations in previous field seasons. The dry spell during all of the 1986 growing season did suppress amoeba populations.

#5. Data analysis.

Synopsis. Statistical analyses mentioned earlier are summarized here. For amoeba counts in soil, by soil dilution procedures, a one-way analysis of variance with 8 replicates per cell was adequate. One-way analysis of variance was used for soil counts (Table 4B) and soil moisture (Table 5) because it is not possible to compare soil horizons or sampling dates. The lower, mineral horizon is roughly twice as dense as the upper, organic horizon so that number of amoebae/g soil and soil moisture differs between horizons. Likewise differences between sampling dates preclude similar comparisons. Growth measurements in culture chambers were analyzed with regression lines, comparing slopes with confidence intervals (a t-test). Other statistical comparisons (e.g. soil chemistry, soil pH, etc.) are done by analysis of variance. For isoenzyme determinations the mainframe computer is needed to do the calculations by Nei's method (Nei, 1972).

SCHEDULE OF WORK ELEMENTS (Nov. 1 to Oct 31 each year)

Element	MONTH 1	2	3	4	5	6	7	8	9	10	11	12
0						X	X	X				
1	X	X	X	X	X	X	X	X	X	X	X	X
2						X	X	X	X	X	X	
3						X	X	X	X	X	X	
4						X	X	X	X	X	X	
5	X	X	X	X	X							
Reports	X	X	X	X	X	X	X	X	X	X	X	X

EXPERIMENTAL

Methods and results will be presented in reference to the Work Plan, given above.

#0. Plot selection and characterization. Site selection is now complete.

Since there was no significant difference in total amoeba numbers between sites, soil characteristics were not as critical as they might be if biological differences had been noted. However, a change in soil characteristics in future years could affect soil-dwelling micro-organisms. Table 1 shows the chemical properties of the organic and mineral horizons for the control, antenna and ground wire sites, with replicates. In comparison with the 1985 data, differences exist between sites which might be attributable to the dry season. Long-range trends are appearing in chemical data but I think an additional season of data should be accumulated before a judgement is made. Table 2 demonstrates some significant differences between sites and sampling dates. Table 3 demonstrates the slightly acidic nature of the soil in a northern hardwood forest, without significant differences between horizons, sites or sampling dates. Comparing the 1983, 1984, 1985 and 1986 seasons indicates slight differences in acidity, possibly due to operator error.

#1. Species and strain characterization. Species of soil amoebae present at the study sites were isolated from soil enrichment plates. So far no species differences have been noted between sites; species composition was the same as in the 1984 field season. Species included Acanthamoeba castellanii, A. polyphaga, A. astronyxis (small strain), Hartmannella sp., Rosculus sp., Naegleria gruberi, Vahlkampfia sp., and Mayorella sp. For the isoenzyme analysis, I have chosen A. polyphaga. A. polyphaga is no more common in soil isolates than other amoebae but its cyst is very distinctive which makes it easy to pick out from soil dilution, enrichment plates (see #2 below). Isoenzyme analyses of 5 clone isolates from each of the 3 study sites are being done. I am using the same isoenzymes as those used by the American Type Culture Collection (Daggett & Nerad, 1983). As a control, I used a strain of this species from the ATCC in the isoenzyme study. Included in the isoenzymes are some that have been used by others, e.g. Pernin et al., 1985. Isoenzyme patterns of A. polyphaga show differences between isolates consistent with a diploid organism. Nei's method for measuring genetic difference (Nei, 1972) is used in this study. The data from the 1985 season revealed more diversity between clonal isolates of the amoeba than was observed among sexually reproducing animals.

However, it was consistent with the large genetic diversity observed among bacterial populations (e.g. ;Howard et al., 1985; Milkman, 1973; Musser et al., 1986; Selander & Levin, 1980). Genetic recombination is either absent or not detected in amoebae while in bacteria recombination is a rare event. In neither case is it required for reproduction. A paper on this work has been published (Jacobson & Band, 1987). The 1986 season data is still being analyzed, I had trouble with one of the isoenzymes which has just been solved. Data completed for the rest of the isoenzymes indicated a significant drop in genetic diversity at all sites. This genetic bottleneck may be due to the drought which would imply that a significant part of the amoeba population died, leaving survivors with a narrow genetic variability. This supports the sensitivity of isoenzymes to physiological stress in the natural population. If ELF radiation introduces stress, it should be reflected in a reduction of isoenzyme diversity as well. Data analysis for the coming season (1987) will be available at the time of the annual report since all isoenzyme procedures are now perfected.

#2. Population size and activity. From the 1983 field data, the number of replicate samples used to determine population size at each study site (i.e. 10) was more than adequate since the coefficient of variation was <10% of the mean.

For a single site, 10 samples, 1 date and 9 D.F., a significant difference at the 90% probability level would be 1.4 X S.D. (from a power curve); for 8 samples per site this would drop a little to 1.5 to 1.6 X S.D. For the 1984, 1985 and 1986 field seasons I chose to use 8 replicates per site since there was little loss in power between 10 and 8 replicate samples per site. Darbyshire's 96 multiwell adaptation of Sing's soil dilution method (Darbyshire et al. 1974) was used. The results of the 1986 season indicate that the control, antenna and ground wire sites have the same number of amoebae/g soil in both the organic and mineral horizons (Table 4, 4B and Figs 1 to 5, 15, 16, & 17). Data on the distribution of amoebae between the cyst and vegetative stages indicates that a proportion of amoebae are in the dormant state for much of the season and vegetative amoeba distribution does differ between sites (Table 4A, 4B and Figs. 1 to 3 and 6 to 9). Specifically, Table 4 gives total counts of vegetative amoebae and cysts while Table 4A gives counts of cysts alone, thus the mathematical difference gives the number of vegetative amoebae present in a sample. Figs. 1, 2 and 3 represent Tables 4 and 4A in showing total counts and cyst counts by horizon and site at various sampling dates. Fig. 4 and 5 compare total counts by horizon, which do not differ significantly (Table 4 and 4B).

The mathematically calculated number of vegetative amoebae is given in Figs. 6 & 7, while the percent vegetative amoebae is given in Figs. 8 & 9. Total comparisons are shown in Fig. 15, 16, & 17. The 1986 season was dry, according to the Climatological Data publications of the Climatic Data Center, NOAA, May through August were all below normal rainfall months (Fig. 14). This is reflected in the small number of amoebae observed in 1986; very atypical in comparison with previous years. A small increase in amoebae was noted at all sites for the July 23 (day 53) count (Table 4, Fig. 17).

#3. Growth and feeding activity. In 1984 I did do growth rate experiments in the soil submersible culture vessels that will be used when the antenna is operational. In 1985 it was not possible to test procedures under the Wisconsin antenna, but this was done in 1986. The data are shown in Tables 6 and 7, and the setup methods area given in the appendix. Table 6 demonstrates that there was no significant difference in growth rate between duplicate culture vessels exposed to voltage and current densities vs. control cultures. Table 7 gives the voltages and current densities measured in the growth chambers. The current density cultures, J-1 & J-2, match the current density in soil while the E-field voltage in culture tubes matched the E-field of soil. In current density cultures the E-field voltage was less than the voltage in soil and in E-field cultures the current density was more than that in soil.

Plans for the 1987 field season: Since the Michigan antenna will be partially operated at reduced power sometime this year, I plan to conduct growth experiments at Michigan facility. Even if there is insufficient power for doing full experiments it will help in developing procedures and logistics needed for in situ experiments.

#4. Ambient monitoring. Table 5 (and Figs. 10 & 11) gives the mean % (w/w) moisture for individual measurements, taken when the soil was sampled, for each set of 8 replicate samples per horizon/site/date. During the growing season (i.e. June, July and August) the soil was drier than in previous years. This correlates to the small number of amoebae observed in soil. However, the soil was not too dry, especially the mineral horizon, in comparison with previous years. Thus the small number of amoebae observed in 1986 correlates in general to a poor growing year, but this may reflect a nutrient limitation rather than an absolute moisture effect. If 1987 also exhibits a drought, I plan to artificially water (i.e. approx. 1 inch/week) a small plot near the control site to see if this supports growth similar to previous growing seasons.

Temperature recordings for the season were somewhat warmer than the 1985 season, consistent with poor tree foliage and drier soil (Figs 12 & 13).

7. Peer reviewers:

I plan to use the following individuals as peer reviewers:

a. Prof. Thomas J. Byers
   Department of Microbiology
   Ohio State University

b. Prof. Fredrick L. Schuster
   Department of Biology
   Brooklyn College

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TABLE 1. SOIL CHEMISTRY:*

ELEM.	DATE***	CO	AO	GO	CM	AM	GM
		SITE/ HORIZON**					
P	1	46,45	40,41	35,36	80,84	22,23	25,28
	2	39,45	41,39	38,35	91,89	23,22	21,21
K	1	108,108	116,124	124,128	36,36	32,32	32,32
	2	84,92	100,104	100,104	36,36	36,32	33,36
Ca	1	2280,2360	2600,2560	2840,2480	632,632	920,800	758,716
	2	2080,2400	2560,2480	2360,2440	840,840	840,716	920,840
Mg	1	160,156	160,188	196,192	63,59	92,76	68,68
	2	120,132	164,176	160,176	84,72	104,80	84,68
NO_3	1	0,0	0,7.4	0,0	3.8,4.4	3.7,4.4	3.6,3.6
	2	9.4,9.0	3.6,4.4	0,0	4.0,4.0	4.3,5.4	4.0,4.9
%Org.N.	1	6.6,5.8	6.6,6.8	5.6,7.2	0.8,0.9	1.2,1.2	1.1,1.2
	2	6.6,6.4	7.2,5.8	6.2,6.2	1.1,0.9	1.1,1.2	1.2,1.2

*Performed by Michigan State University Soil Testing Laboratory, data expressed as ppm except for %org.N.

**SITE: C, control; A, antenna; G, ground.
HORIZON: O, organic; M, mineral

***Data was obtained June 15 and July 23, 1986, each of which were taken from 20 random samples.

TABLE 2. SOIL CHEMISTRY 2X ANOVA: Two-way analysis of variance between sites /dates:

ELEMENT		ORGANIC			MINERAL	
		D.F.	M.S.	F	M.S.	F
P	Site	2	60.25	13.904 **	5272.58	2041 **
	Date	1	2.9999	0.6923 NS	2.08334	0.8065 NS
	Interact.	2	5.25	1.2115 NS	46.0833	17.839 **
	Error	6	4.3333	----	2.58333	----
K	Site	2	289.33	19.727 **	11.0833	5.32 *
	Date	1	1281.3	87.364 **	6.7499	3.24 NS
	Interact.	2	9.3333	0.6364 NS	1.75	0.84 NS
	Error	6	14.666	----	2.083	----
Ca	Site	2	90533.3	4.2975 NS	8170.34	2.5842 NS
	Date	1	53333.4	2.5316 NS	24120.33	7.629 *
	Interact.	2	12133.3	0.5759 NS	23158.33	7.3247 *
	Error	6	21066.6	----	3161.666	----
Mg	Site	2	1668	14.717 **	403	3.875 NS
	Date	1	1281.33	11.306 *	363	3.4904 NS
	Interact.	2	217.333	1.9176 NS	26.9999	0.2596 NS
	Error	6	113.333	----	104	----
NO_3	Site	2	24.3633	5.2621 *	0.2275	0.9512 NS
	Date	1	30.0833	6.4975 *	0.8008	3.3484 NS
	Interact.	2	27.3233	5.9014 *	0.2858	1.1951 NS
	Error	6	4.63	----	0.2392	----
%Org N	Site	2	0.10333	0.2366 NS	0.08333	14.286 **
	Date	1	0.00333	0.0076 NS	0.0075	1.2857 NS
	Interact.	2	0.08333	0.1908 NS	0.009999	1.7143 NS
	Error	6	0.43666	----	0.005833	----

* = 5% significance level
** = 1% significance level

TABLE 3. SOIL pH:

DATE	SITE	HORIZON	MEAN pH ± S.D. (n=10)
26JUN86	Control	Organic	6.42 ± 0.49
		Mineral	6.5 ± 0.43
	Antenna	Organic	6.59 ± 0.41
		Mineral	6.60 ± 0.29
	Ground	Organic	6.24 ± 0.32
		Mineral	6.24 ± 0.35
30JULY86	Control	Organic	6.4 ± 0.54
		Mineral	6.6 ± 0.77
	Antenna	Organic	6.69 ± 0.32
		Mineral	6.65 ± 0.21
	Ground	Organic	6.7 ± 0.41
		Mineral	6.21 ± 0.34

Three-way ANOVA:

	D.F.	M.S.
#1. Site	2	1.7121
#2. Horizon	1	0.0608
#3. Date	1	0.3968
#4. Site X Horizon	2	0.4067
#5. Site X Date	2	0.1022
#6. Horizon X Date	1	0.1267
#7. Site X Horizon X Date	2	0.2223
Error	108	0.1854

F-TESTS (none signif.):

Test	F
1/7	3.8518
2/7	0.2734
3/7	1.7851
4/7	1.8301
5/7	0.46
6/7	0.5702

Table 4. Total counts from 8 samples per horizon/site:

SITE	HORIZON	DATE	MEAN #/g soil* ± S.E.	MEAN** (#/g soil)
Control	Organic	6/16	7.4491 ± 0.1657	1868
		7/23	8.8460 ± 0.2632	8712
		8/21	7.5659 ± 0.1633	2119
		9/13	7.9171 ± 0.0963	2840
		10/14	8.1682 ± 0.0963	3650
	Mineral	6/16	6.7327 ± 0.1132	878
		7/23	7.7145 ± 0.1419	2394
		8/21	7.2806 ± 0.0783	1482
		9/13	6.9847 ± 0.1227	1136
		10/14	7.1285 ± 0.1226	1311
Antenna	Organic	6/16	7.3258 ± 0.1381	1631
		7/23	9.5938 ± 0.2529	18313
		8/21	7.9847 ± 0.2030	3466
		9/13	8.0000 ± 0.0898	3062
		10/14	7.9502 ± 0.1383	3944
	Mineral	6/16	6.3888 ± 0.0671	605
		7/23	7.6807 ± 0.1761	2410
		8/21	7.3713 ± 0.1156	1667
		9/13	7.3355 ± 0.1259	1613
		10/14	7.4924 ± 0.1156	1882
Ground	Organic	6/16	6.9676 ± 0.1057	1100
		7/23	9.1213 ± 0.2975	12821
		8/21	7.7325 ± 0.1937	2613
		9/13	7.6329 ± 0.1753	2268
		10/14	7.9886 ± 0.1788	3303
	Mineral	6/16	6.5730 ± 0.1346	763
		7/23	8.1089 ± 0.1757	3698
		8/21	7.3121 ± 0.1427	1610
		9/13	7.0862 ± 0.1411	1271
		10/14	7.1169 ± 0.1213	1288

* Mean expressed as the natural log of amoeba number, used to calculate analysis of variance (Table 4B).

** The mean calculated from log transformed data and the mean calculated from the original arithmatic data cannot be interchanged. Computer calculations round off log transformations which give rise to this error. In the 1985 report, the arithmatic mean was calculated from the mean of the log transformed data.

Table 4A. Cyst counts from 8 samples per horizon/site:

SITE	HORIZON	DATE	MEAN #/g soil* ± S.E.	MEAN (#/g soil)
Control	Organic	6/16	7.1908 ± 0.2354	1487
		7/23	7.2839 ± 0.1623	1627
		8/21	6.9156 ± 0.1352	1085
		9/13	7.1901 ± 0.1225	1405
		10/14	7.4409 ± 0.1224	1905
	Mineral	6/16	6.3835 ± 0.0894	610
		7/23	6.6305 ± 0.4000	762
		8/21	6.6304 ± 0.0709	771
		9/13	6.2266 ± 0.0747	517
		10/14	6.3703 ± 0.0747	597
Antenna	Organic	6/16	6.9142 ± 0.0728	1027
		7/23	7.1947 ± 0.1300	1410
		8/21	6.9985 ± 0.0800	1121
		9/13	7.6631 ± 0.1313	1978
		10/14	7.1277 ± 0.0801	1275
	Mineral	6/16	6.2918 ± 0.0749	552
		7/23	6.6080 ± 0.0651	753
		8/21	6.3414 ± 0.0278	569
		9/13	6.2396 ± 0.0656	521
		10/14	6.4629 ± 0.0278	643
Ground	Organic	6/16	6.7506 ± 0.0627	866
		7/23	7.6662 ± 0.1772	2391
		8/21	7.0387 ± 0.1333	1222
		9/13	7.0186 ± 0.1024	935
		10/14	7.4687 ± 0.1024	1817
	Mineral	6/16	6.2044 ± 0.0805	507
		7/23	6.6672 ± 0.0893	786
		8/21	6.4884 ± 0.0755	671
		9/13	6.4030 ± 0.1184	642
		10/14	6.3783 ± 0.0245	590

* Mean expressed as the natural log of amoeba number, used to calculate analysis of variance (Table 4B).

TABLE 4B. One-way analysis of variance by date and horizon, data transformed to ln (see Table 4 & 4A).

HORIZON	DATE	GROUPS	DF	MS	F
				TOTAL COUNT	
ORGANIC	6/16	among	2	0.479270458	
		within	21	0.142334461	3.36721307 NS
	7/23	among	2	0.501909971	
		within	21	0.199098246	2.52091608 NS
	8/21	among	2	0.355528116	
		within	21	0.281089192	1.26482314 NS
	9/13	among	2	0.272109509	
		within	21	0.1130476	2.40703482 NS
	10/14	among	2	0.104514122	
		within	21	0.155619884	0.67159876 NS
MINERAL	6/16	among	2	0.236403191	
		within	21	0.094525995	2.50622264 NS
	7/23	among	2	0.00713757	
		within	21	0.036843232	0.19372724 NS
	8/21	among	2	0.01697731	
		within	21	0.106292293	0.15972287 NS
	9/13	among	2	0.260635853	
		within	21	0.135443301	1.92431704 NS
	10/14	among	2	0.364740849	
		within	21	0.114912328	3.17407935 NS
				CYST COUNT	
ORGANIC	6/16	among	2	0.396097422	
		within	21	0.103083247	3.84250043 *
	7/23	among	2	1.14410234	
		within	21	0.591390201	1.93459806 NS
	8/21	among	2	0.0315136909	
		within	21	0.113209361	0.27836648 NS
	9/13	among	2	0.89142251	
		within	21	0.113924254	7.84246948 **
	10/14	among	2	0.286993504	
		within	21	0.0850521723	3.37432303 NS
MINERAL	6/16	among	2	0.0641460419	
		within	21	0.0535706793	1.19740953 NS
	7/23	among	2	0.453520298	
		within	21	0.218675909	2.0739381 NS
	8/21	among	2	0.166992784	
		within	21	0.0306793168	5.44317153 *
	9/13	among	2	0.0773634911	
		within	21	0.0637202717	1.21411113 NS
	10/14	among	2	0.0207060575	
		within	21	0.0190404415	1.08747781 NS

* = 5% significance level
** = 1% significance level

TABLE 5. SOIL MOISTURE (%, w/w)1:

	CONTROL SITE		ANTENNA SITE		GROUND SITE	
HORIZON:	ORG	MIN	ORG	MIN	ORG	MIN
DATE:						
6/16	38.5±9.7	14.8±1.9	35.5±11	12.2±2.4	27.3±6.8	13.1±1.9
7/23	17.3±3.3	8.6±2.9	34.7±9.3	8.6±3	33.7±7.7	10 ±2
8/21	27.5±10	10.6±2.9	34.7±12	8.9±3.7	38.6±8.3	10.5±2
9/13	43 ±15	16.9±3.6	55.9±3.5	15.3±3.7	34.5±10.3	14.2±1.5
10/14	55.7±8.1	23±3.8	42.6±7	19.3±4.7	58.3±7.6	21.8±3

ONE-WAY ANOVA:

		ORGANIC		MINERAL	
Date		D.F.	M.S.	D.F.	M.S.
6/16	Between	2	325.731911	2	27.148552
	Within	21	70.660251	21	91.748976
	F=	4.6098324 *		3.1069534 NS	
7/23	Between	2	677.493515	2	6.016663
	Within	21	52.353803	21	7.250587
	F=	12.9406743 **		0.8298174 NS	
8/21	Between	2	374.934845	2	0.420549
	Within	21	85.708315	21	4.641707
	F=	4.3745445 *		0.09060241 NS	
9/13	Between	2	873.604965	2	13.231162
	Within	21	117.445924	21	9.444602
	F=	7.4385914 **		1.40097274 NS	
10/14	Between	2	476.271584	2	28.805769
	Within	21	55.388226	21	13.687277
	F=	8.5987874 **		2.10456531 NS	

1 Sample size, 8

* = 5% significance level
**= 1% significance level

Table 6. Regression calculations for growth of _Acanthamoeba polyphaga_, log transformed, in soil submerged culture vessels at the Wisconsin transmitter facility. Correlation coefficient also shown, as a separate calculation to indicate the distribution of data.

Experiment	Slope*	95% Confidence Limits**	Correl. Coeff.
E-field	0.01845	L1 = 0.051837 L2 = 0.088738	0.9578
Current density	0.01211	L1 = 0.055395 L2 = 0.079630	0.9111
Control	0.01247	L1 = 0.036606 L2 = 0.061564	0.8283

* Mean Generation Time = 7 hr.

** For the slope of the growth curve.

TABLE 7. Culture cell current densities and E-field voltages, Wisconsin Transmitter, 1986.

Electrode[1]	date/time	Voc (mv)	Vcl (mv)	Vr (mv)	Ecl[2] (mv/m)	Jcl[3] (mA/m2)
J-1	8/14,0946	1210	0.23	1200	2	2.3
	1429	1210	0.5	1200	4.4	2.3
	8/15,0850	1222	0.61	1210	5.4	2.3
J-2	8/14,0948	1215	0.23	1210	2	2.3
	1432	1215	0.32	1210	2.3	2.3
	8/15,0852	1220	0.35	1215	3.1	2.3
E-3	8/14,0950	1920	158	13.5	1398	639
	1435	1920	171 to 160	12.7	1416	601
	8/15,0855	1920	160	12.7	1416	601
E-4[4]	8/14,0953	1780	370 to 158	9.5	1398	393
	1438	1780	175	11.0	1549	435
	8/15,0859	1780	175	11.0	1549	455

[1] J electrodes: current density; E electrodes: E-fields.

[2] E-field: Ecl(V/m) = Vcl / 0.113m (length between electrodes).

[3] Current density: Jcl (A/m^2) = Vr / R X xs. area of cl (m^2), see #4 below.

where R (ohms) = 2.5×10^6 for J ; 100 for E

[4] Adjusted Vcl by adding more fluid to the culture vessel so that the final volume was 15.6 ml rather than the 7.5 ml used in the other vessels. Submerged electrode area ("area of cl" cited above) was:

a. 7.5 ml = 2.11×10^{-4} m^2

b. 15.6 ml = 2.42×10^{-4} m^2

Figure 1. Control site total counts and cyst counts.

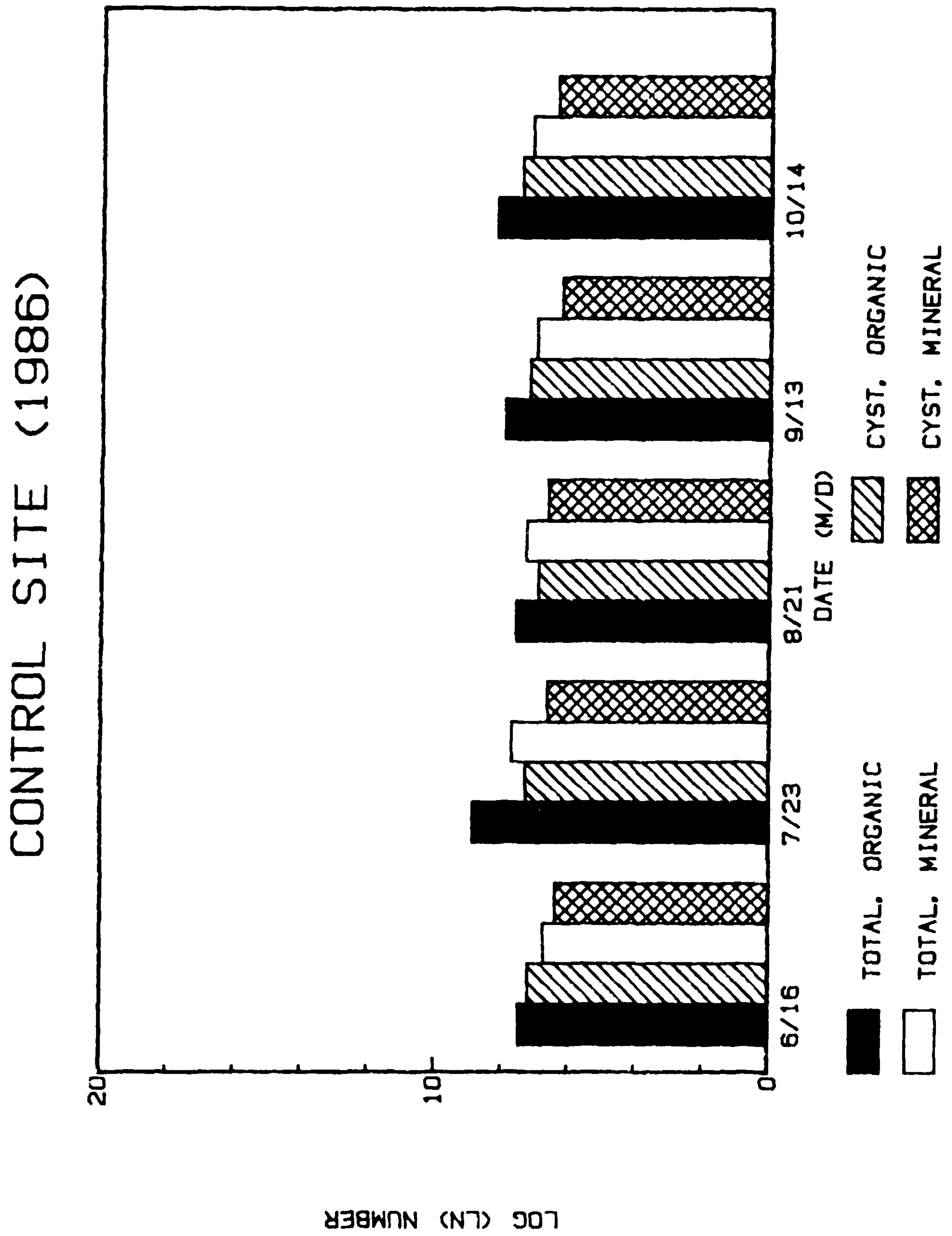


Figure 2. Antenna site total counts and cyst counts.

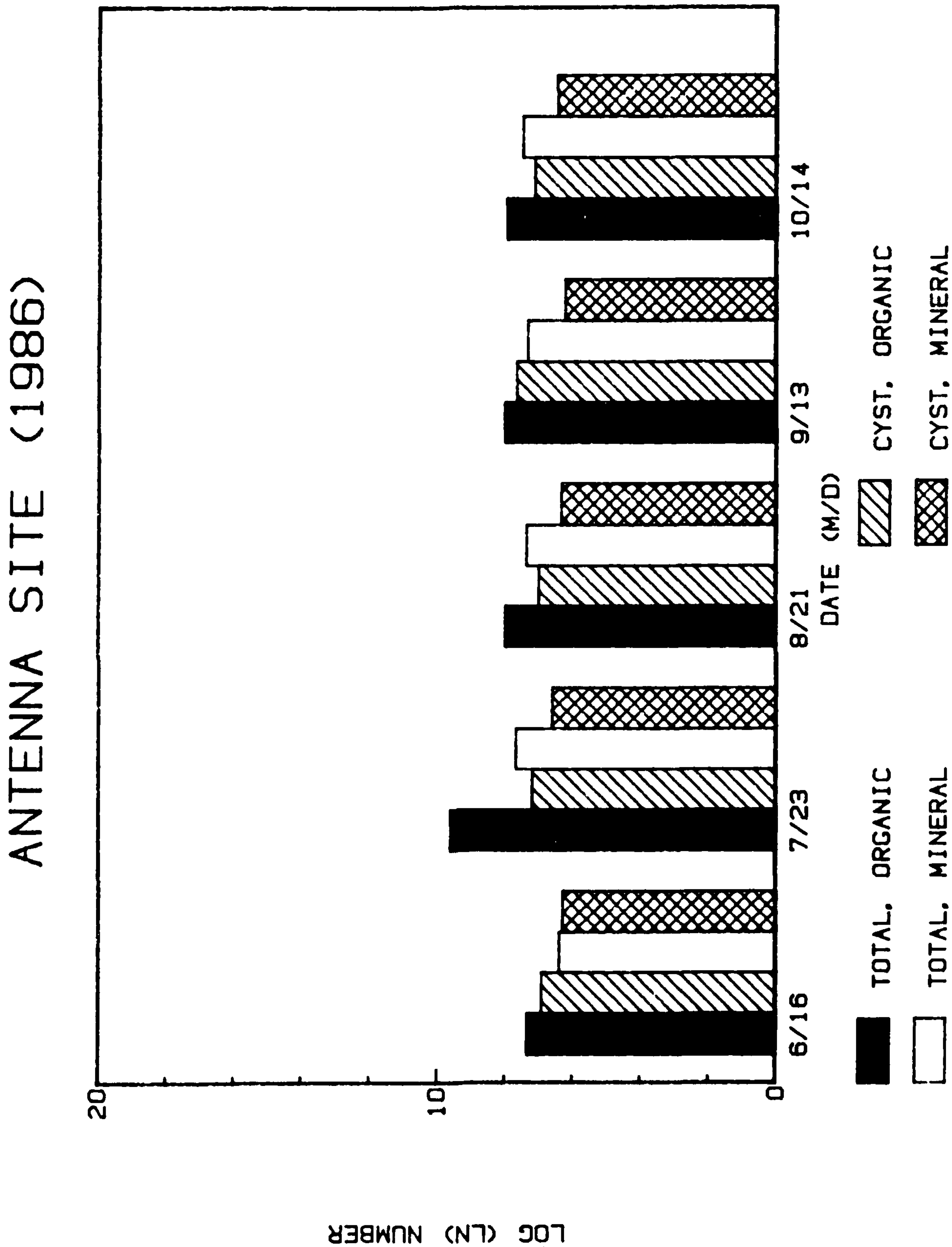


Figure 3. Ground site total counts and cyst counts.

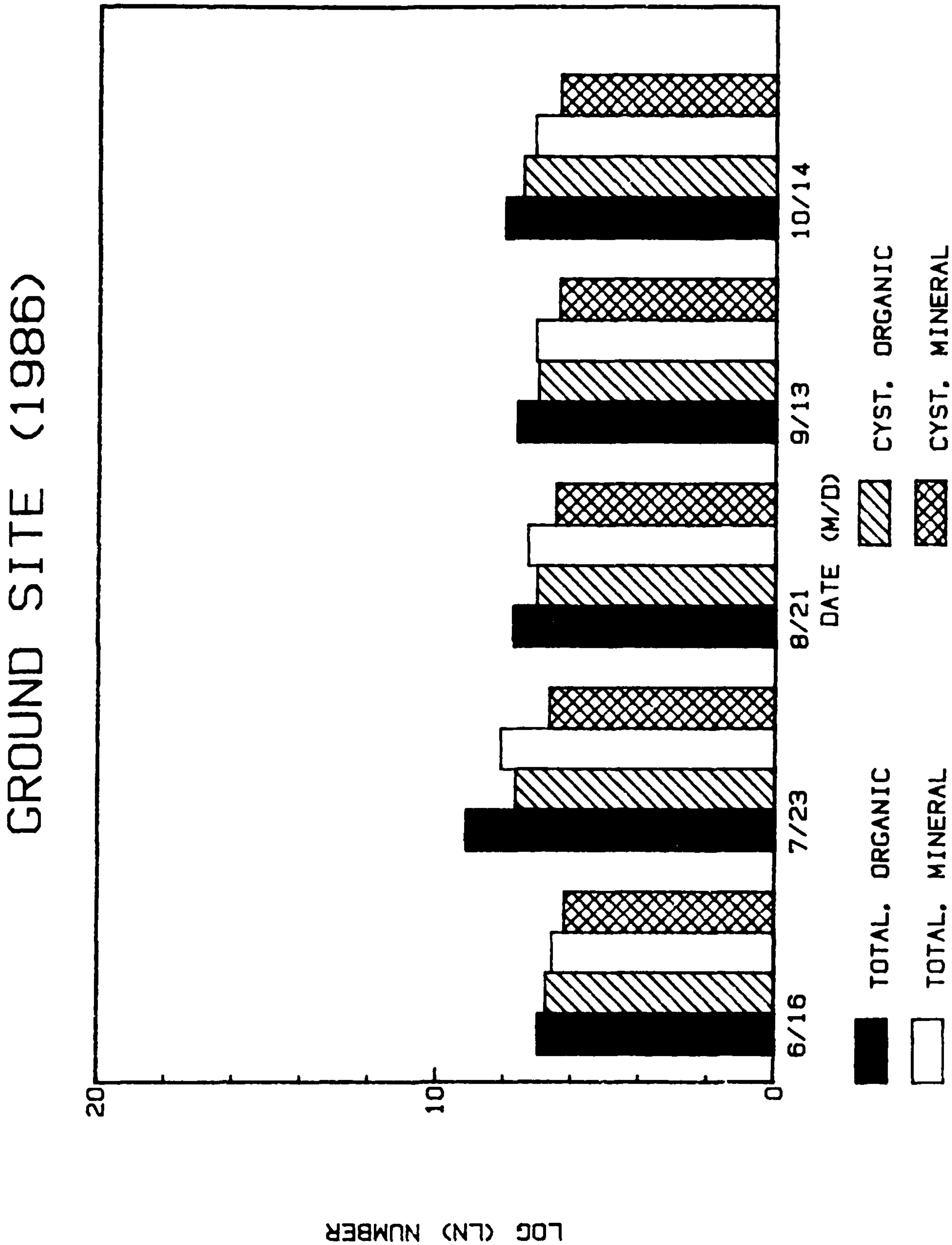


Figure 4. Average total number of amoebae at the 3 sites, ORGANIC HORIZON

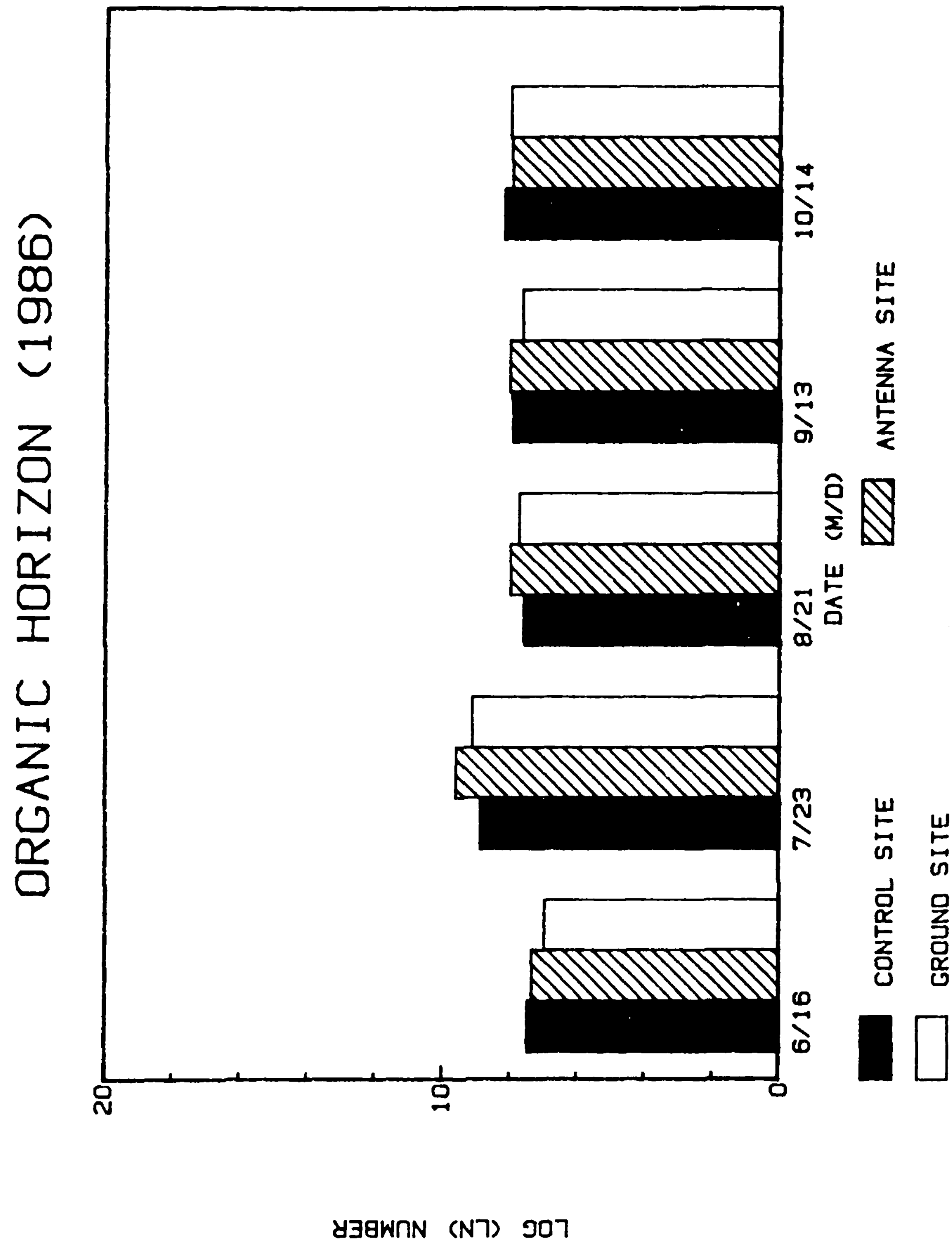


Figure 5. Average number of amoebae at the 3 sites, MINERAL HORIZON

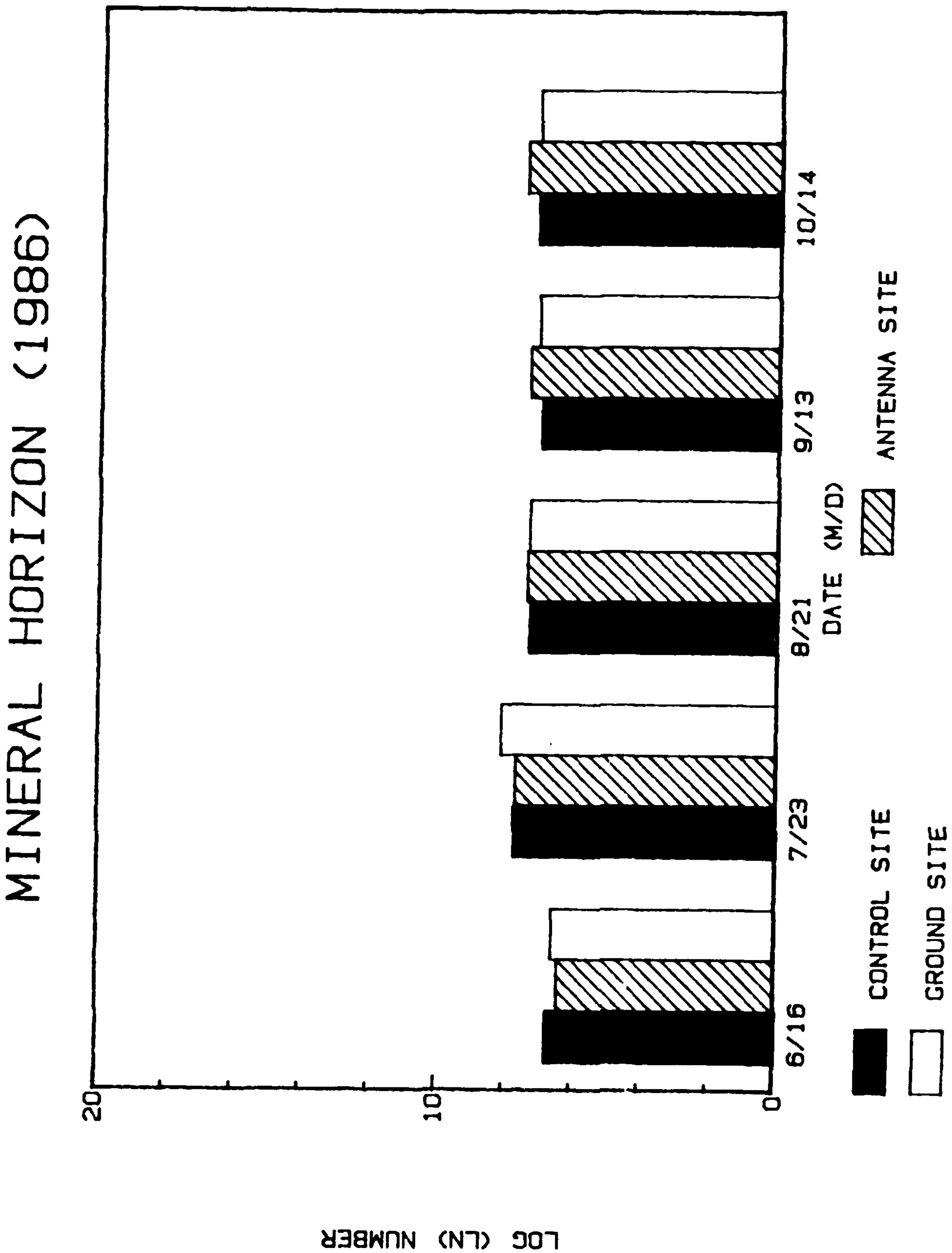


Figure 6. Calculated number of vegetative amoebae from Table 4 and 4A (by subtracting means); plotted to same scale used in 1985.

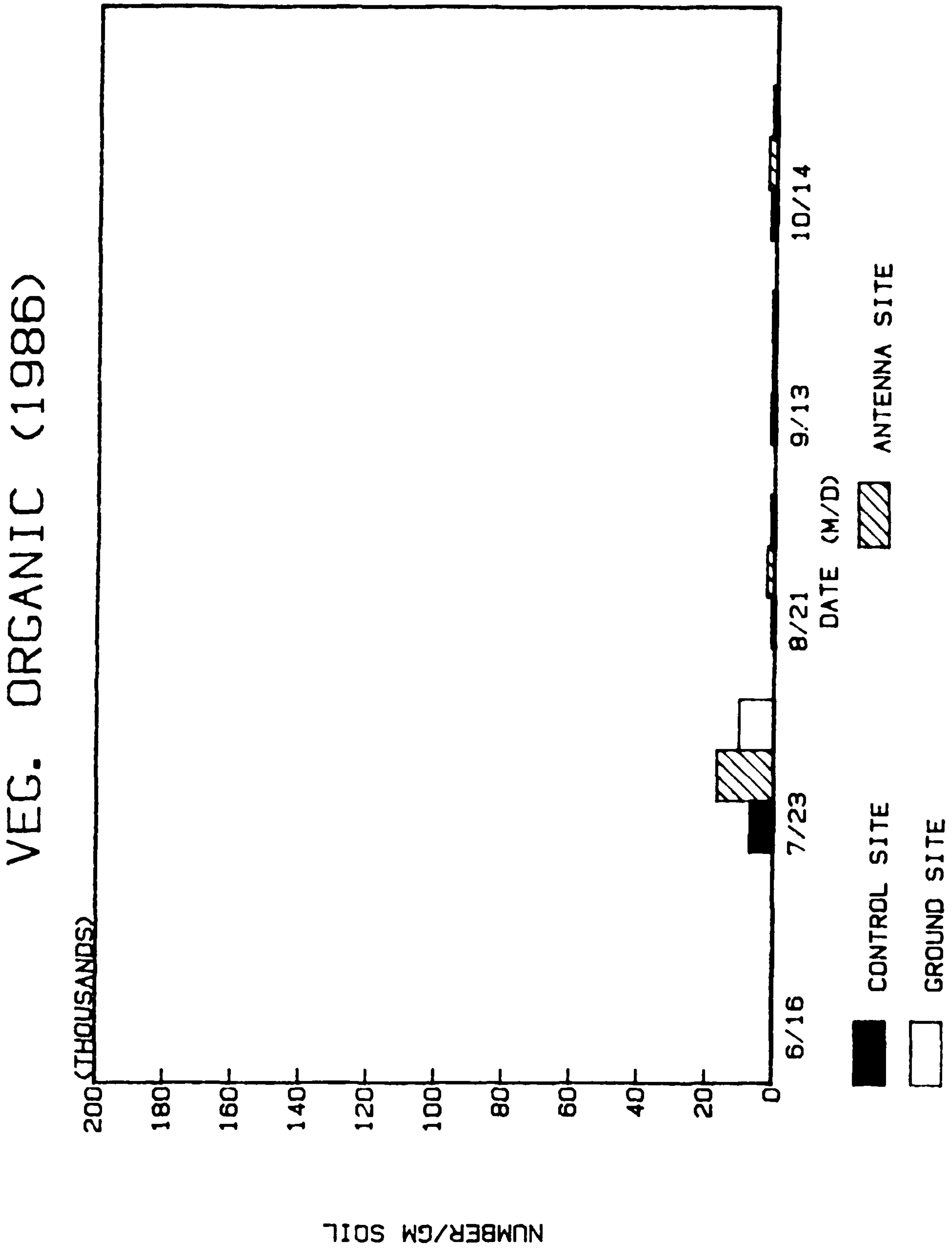


Figure 6A. Figure 6 replotted to larger scale.

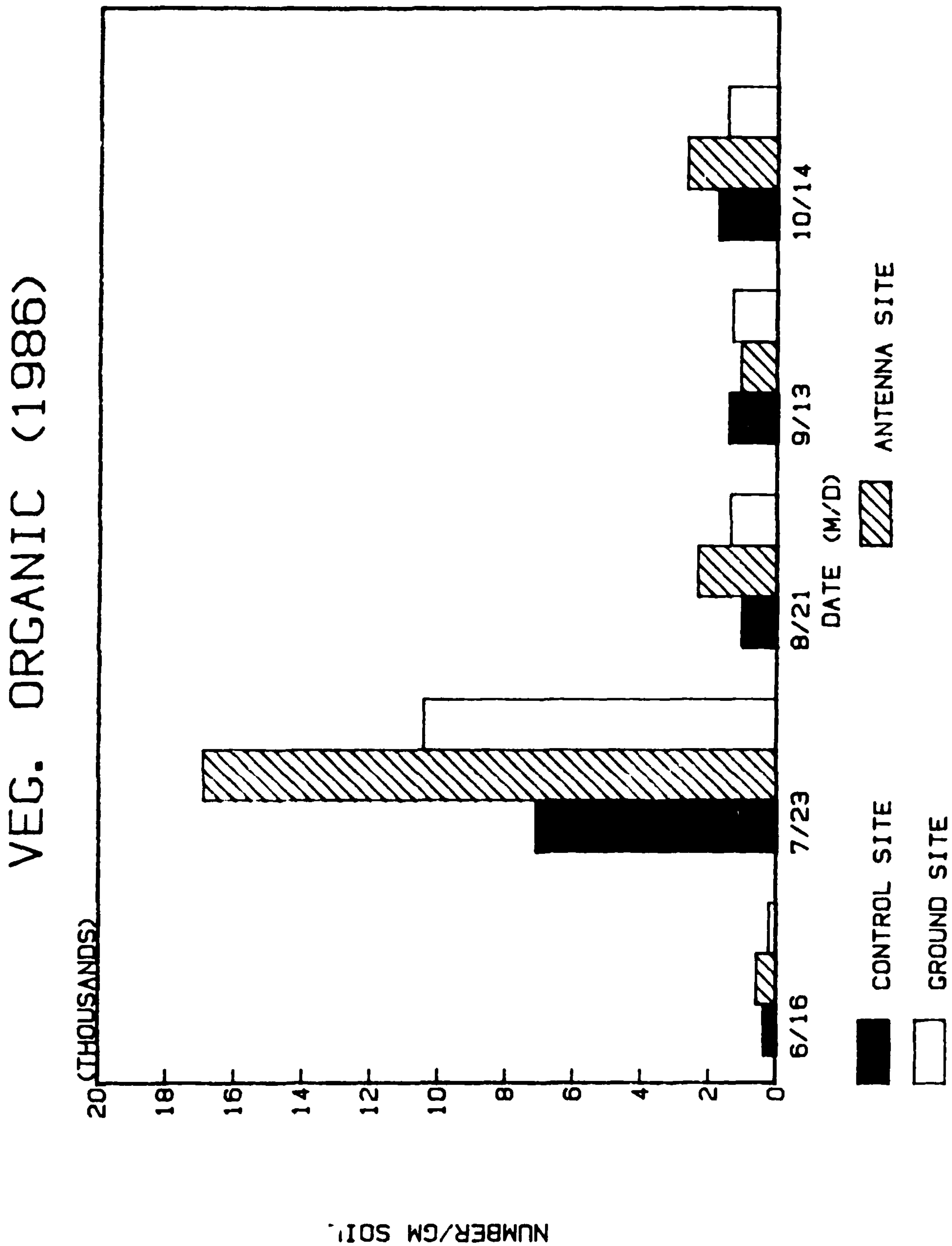


Figure 7. Calculated number of vegetative amoebae from Table 4 and 4A (by subtracting means); plotted to same scale used in 1985

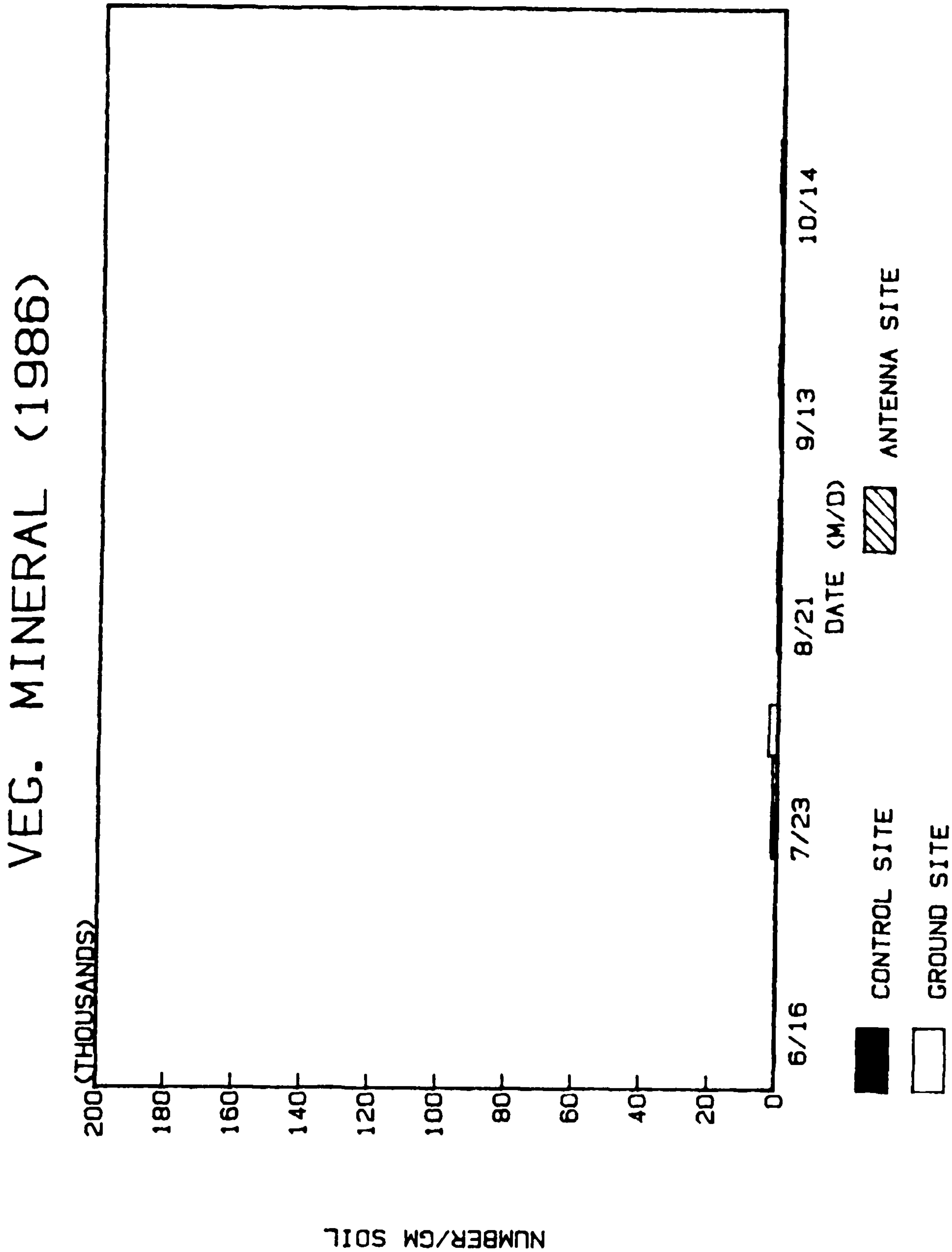


Figure 7A. Figure 7 replotted to larger scale.

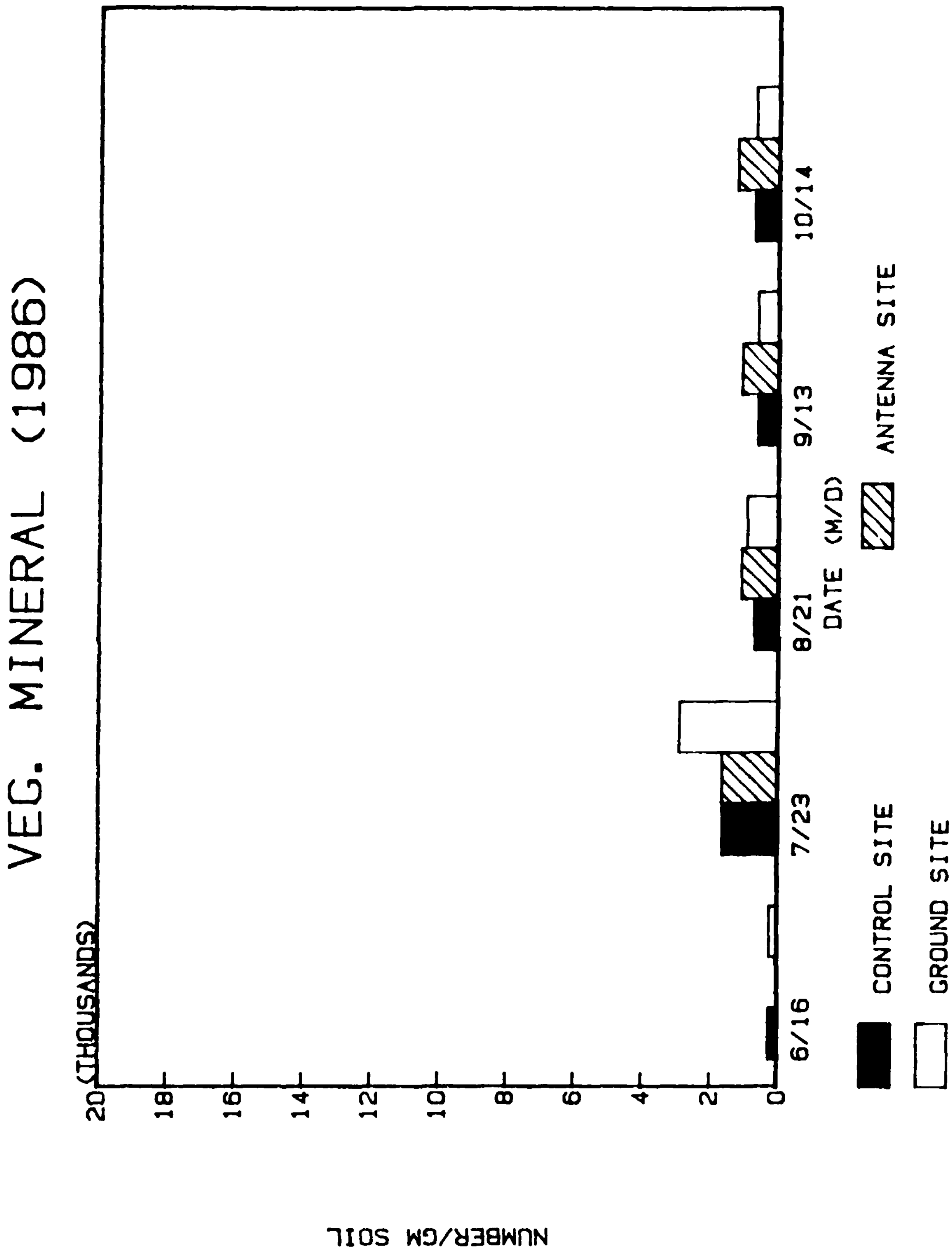


Figure 8. Percent vegetative amoebae, ORGANIC horizon.

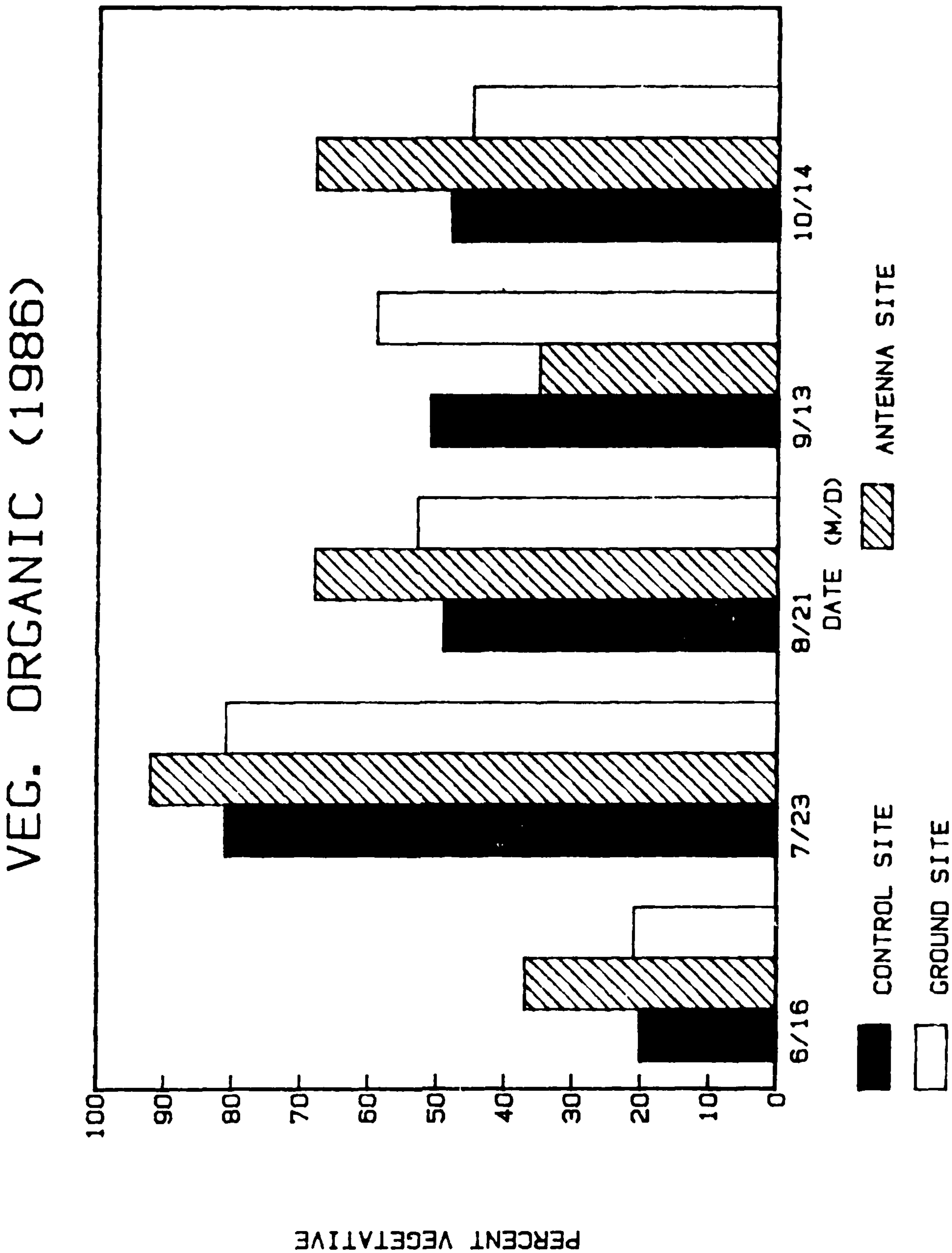


Figure 9. Percent vegetative amoebae, MINERAL horizon.

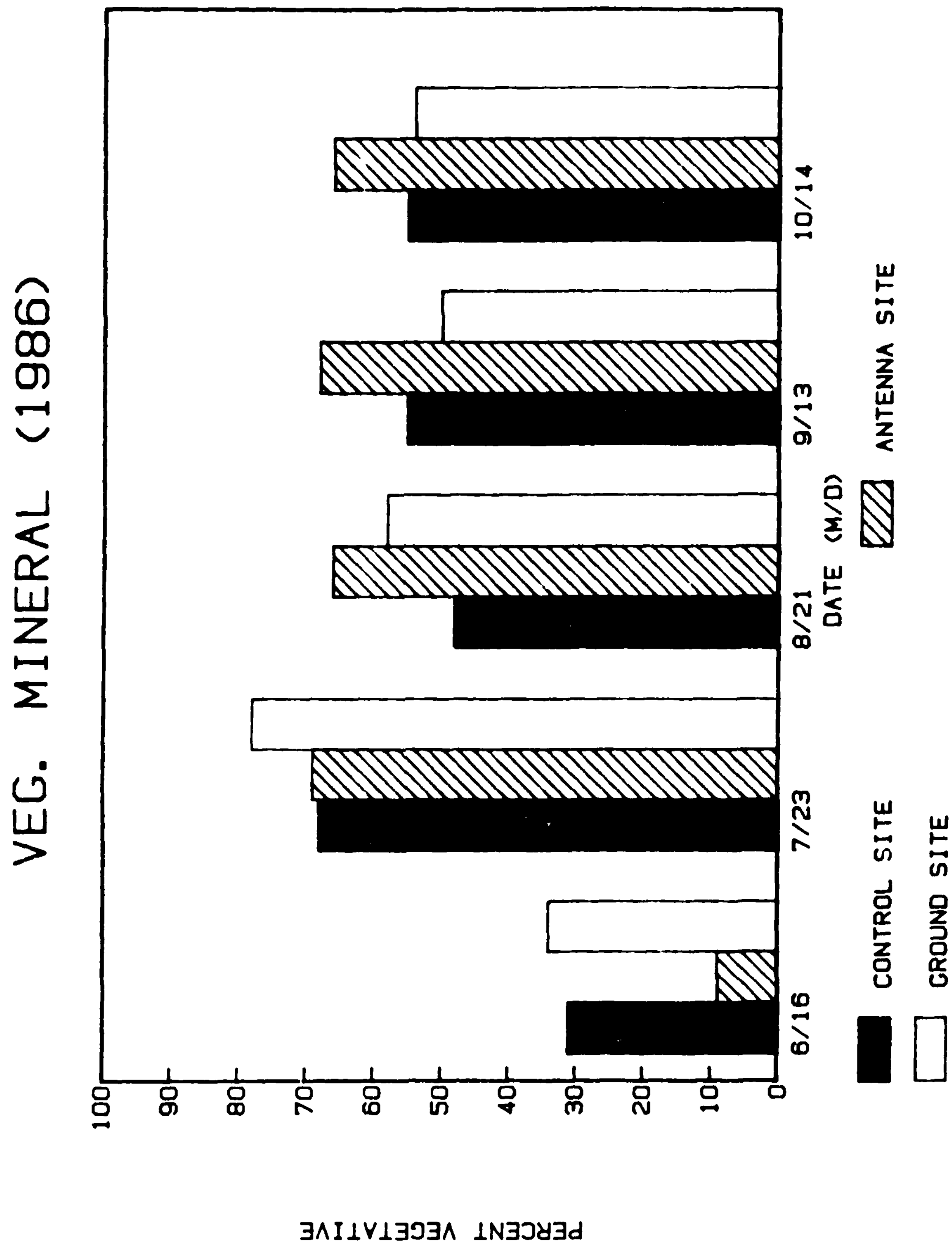


Figure 10. Moisture content of samples taken for counting, ORGANIC horizon

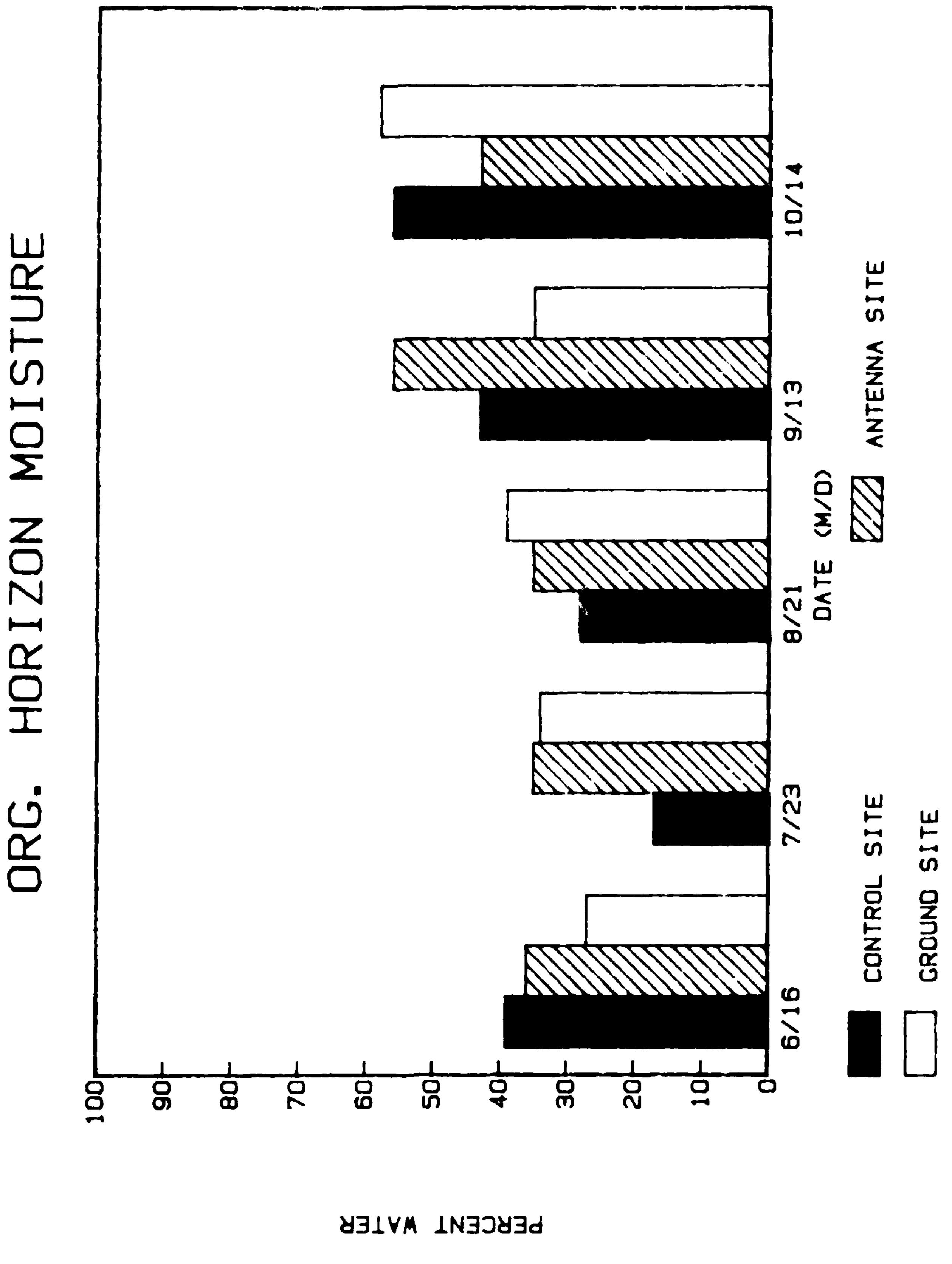


Figure 11. Moisture content of samples taken for counting, MINERAL horizon.

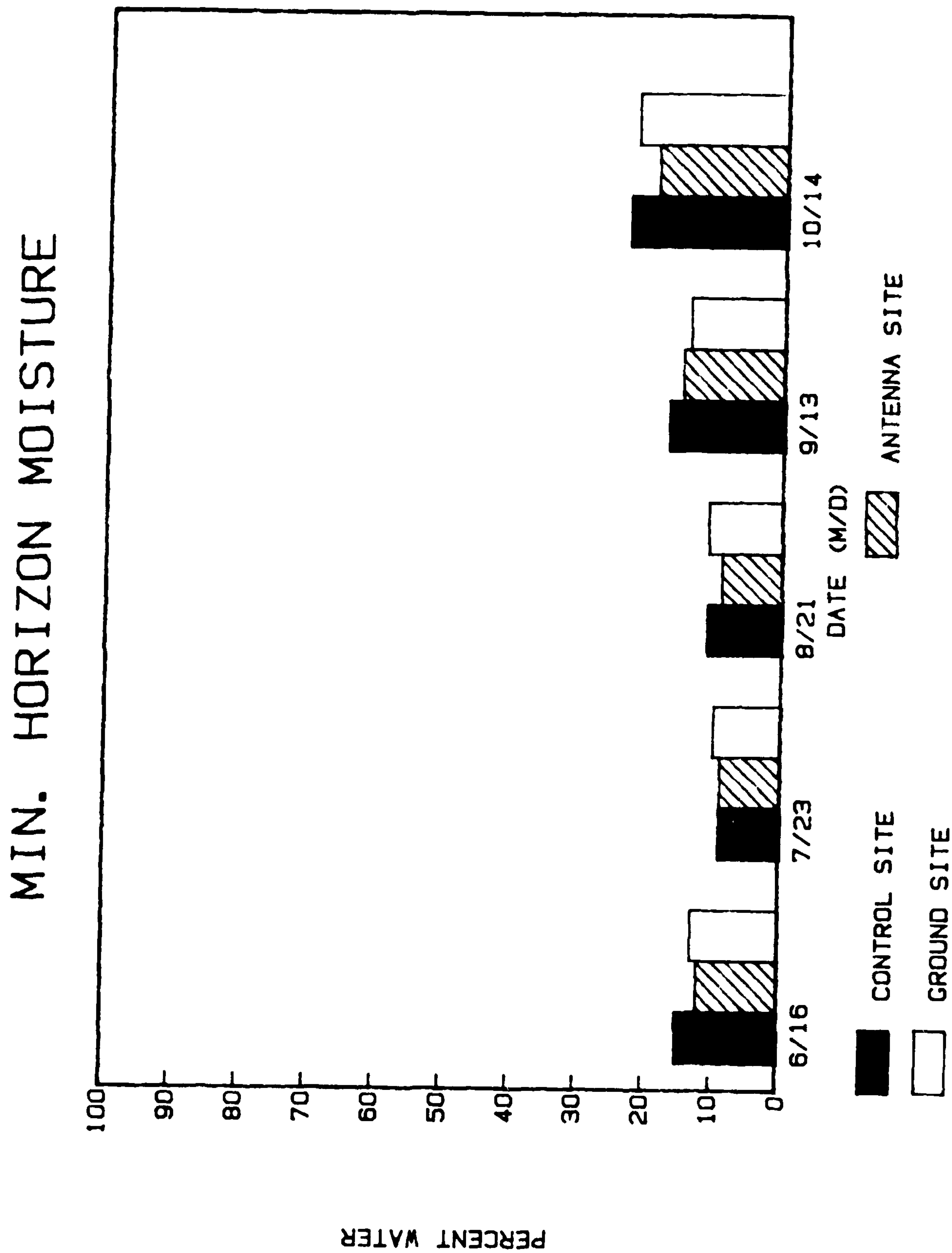


Figure 12. Pooled temperature records, mineral horizon, all sites, mean daily temperature with S.D. error bars. Points plotted every 3rd day.

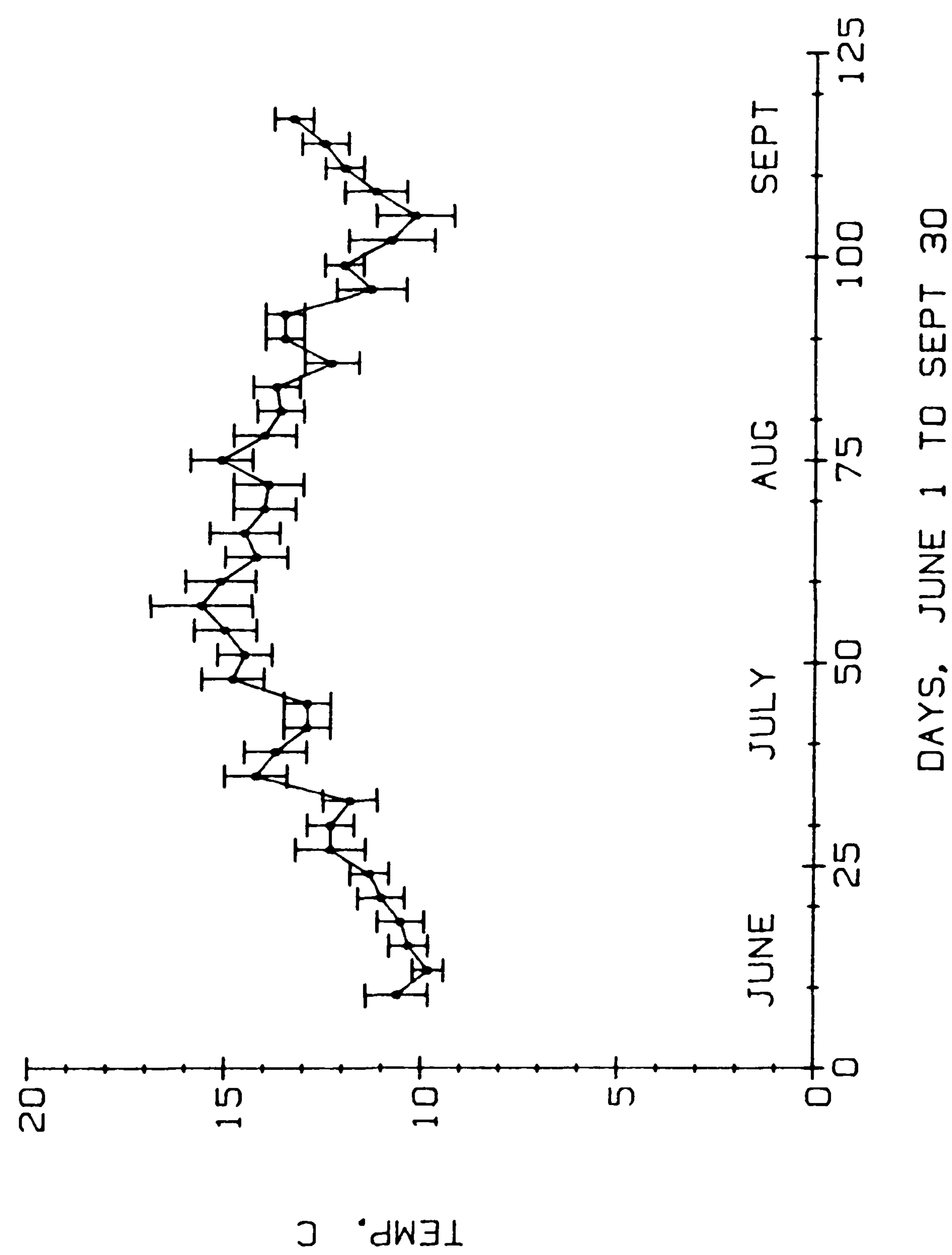


Figure 13. Figure 12 temperature data plotted with the same data from the 1985 season (points as darker, x's); for much of the season, by inspection, the 1986 temperatures were warmer.

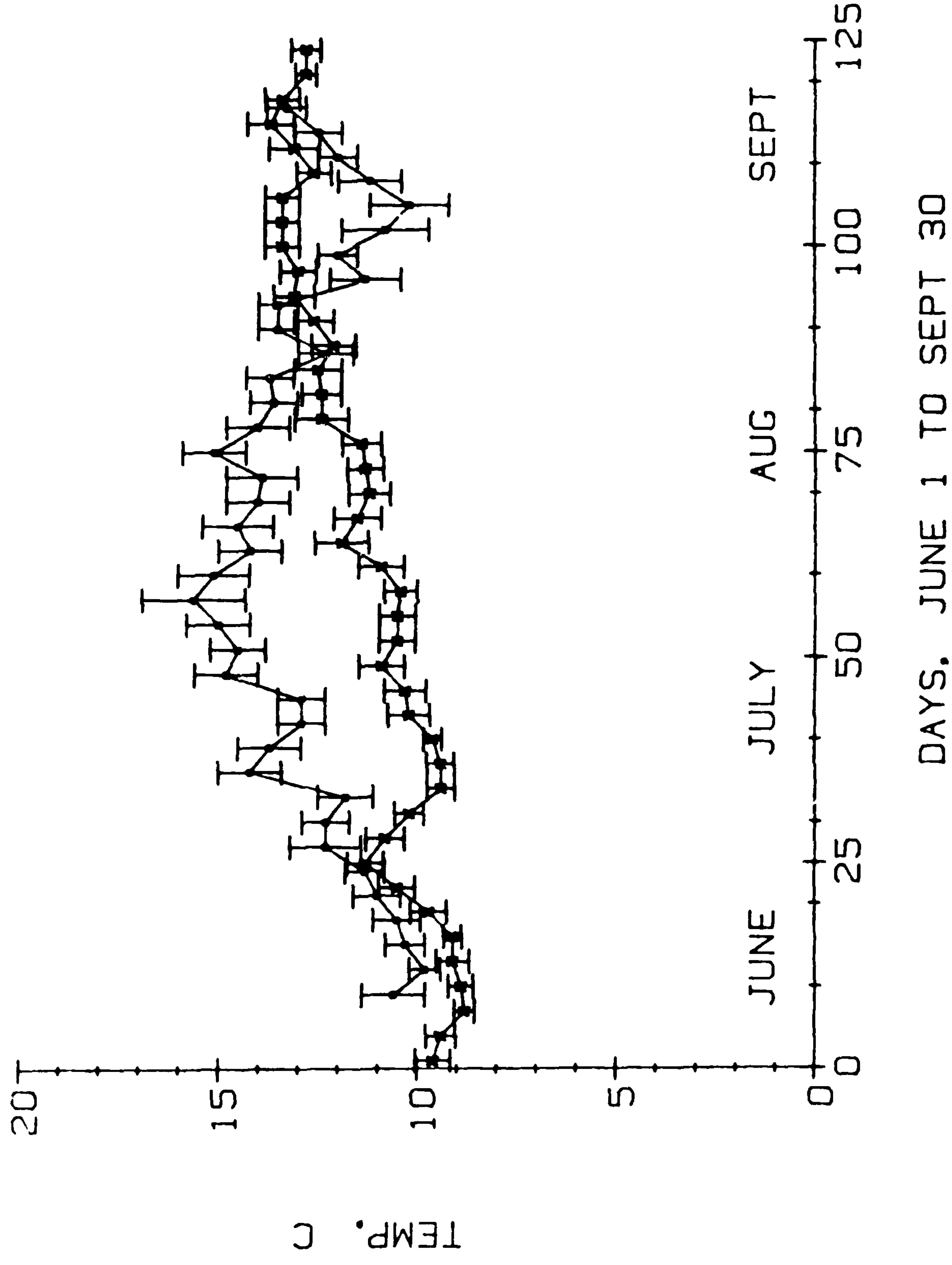


Figure 14. Annual rainfall by month for 3 seasons vs. average rainfall for 1951 to 1980 (i.e. normal). Data exerpted from the Climatological Data for Michigan, published by NOAA.

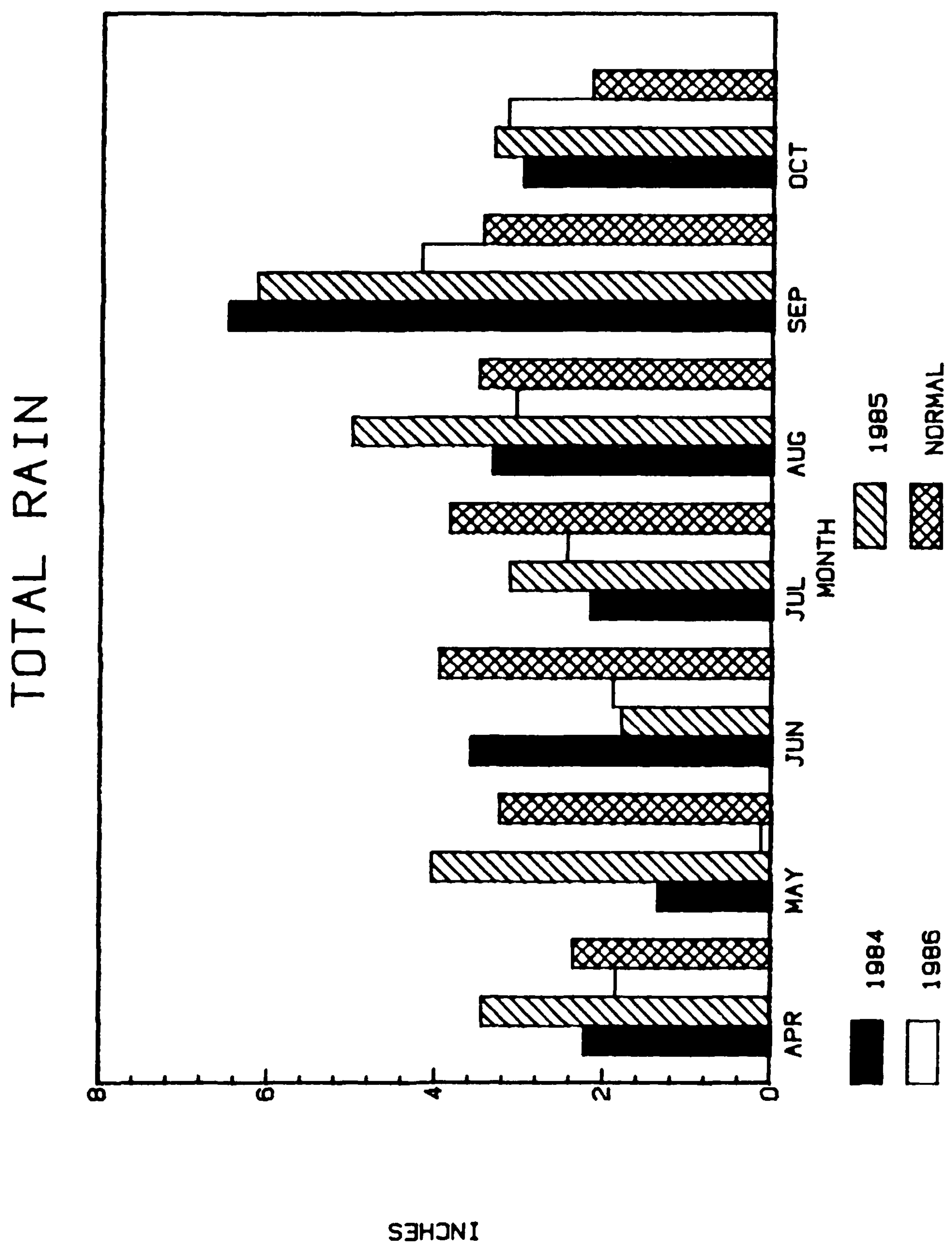


Figure 15. Summary of 1984 amoeba counts, given both as log counts and absolute numbers, % vegetative amoebae and % soil water.

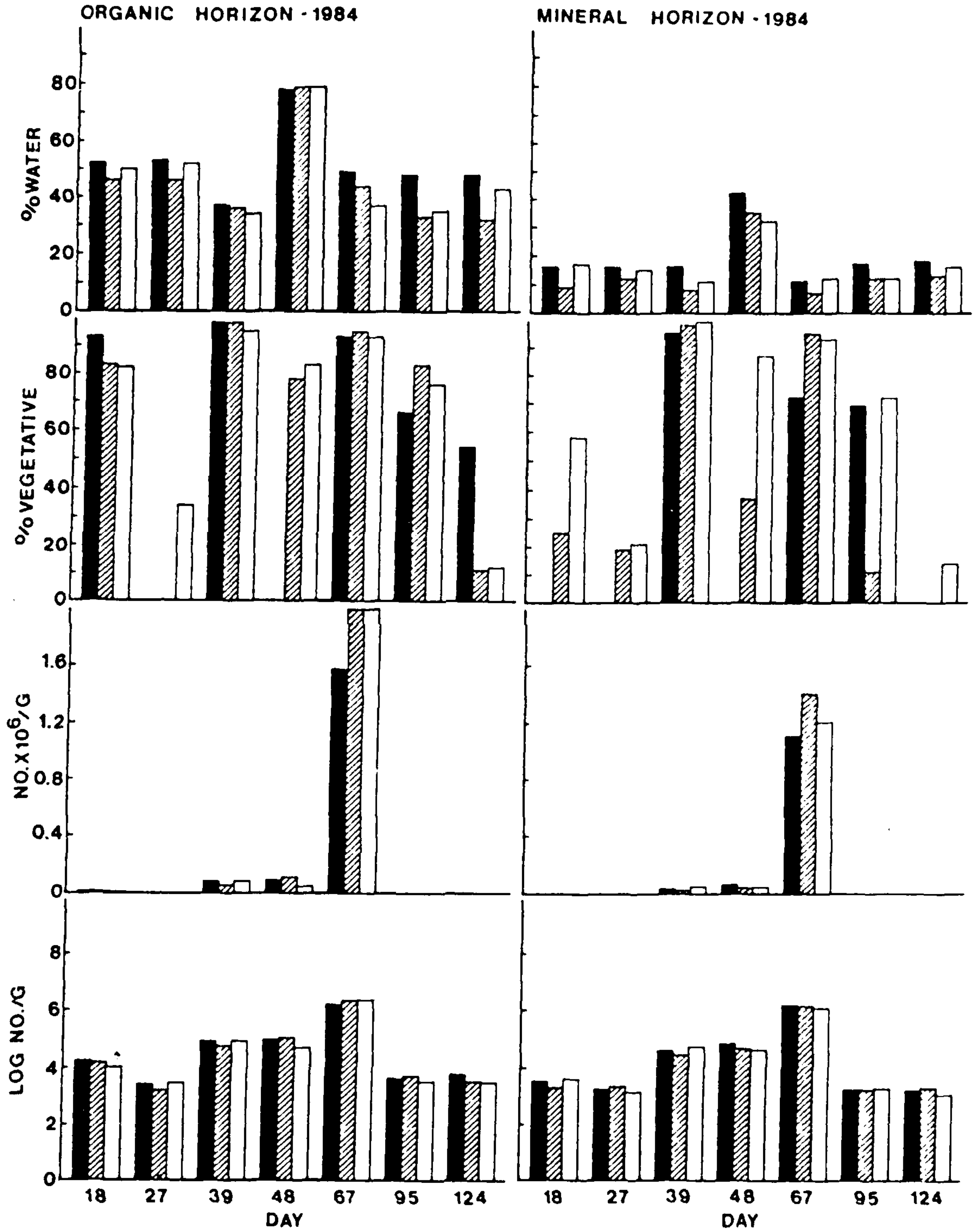


Figure 16. Summary of 1985 amoeba counts, given both as log counts and absolute numbers, % vegetative amoebae and % soil water.

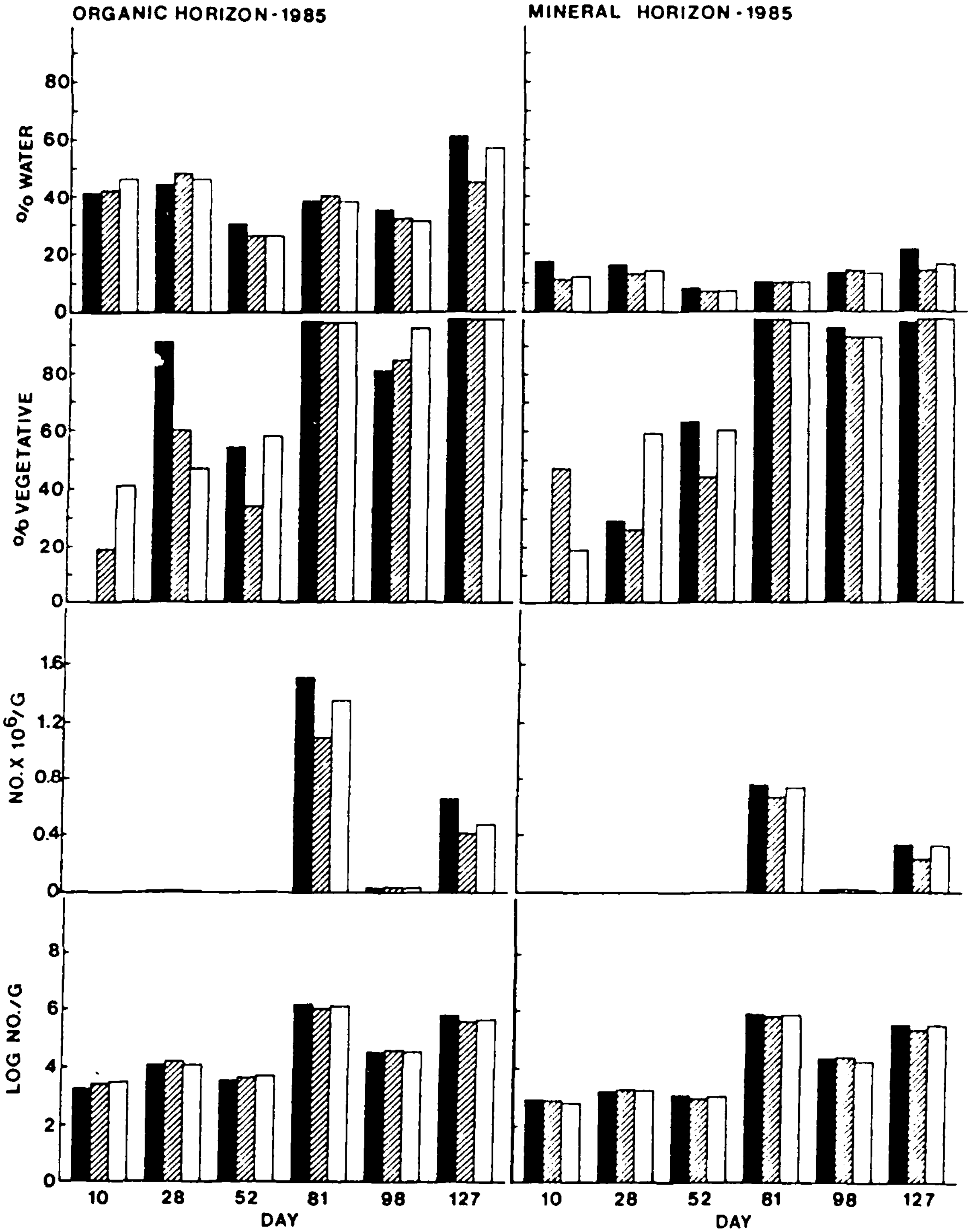


Figure 17. Summary of 1986 amoeba counts, given both as log counts and absolute numbers, % vegetative amoebae and % soil water. Note that the absolute number scale differs from previous years.

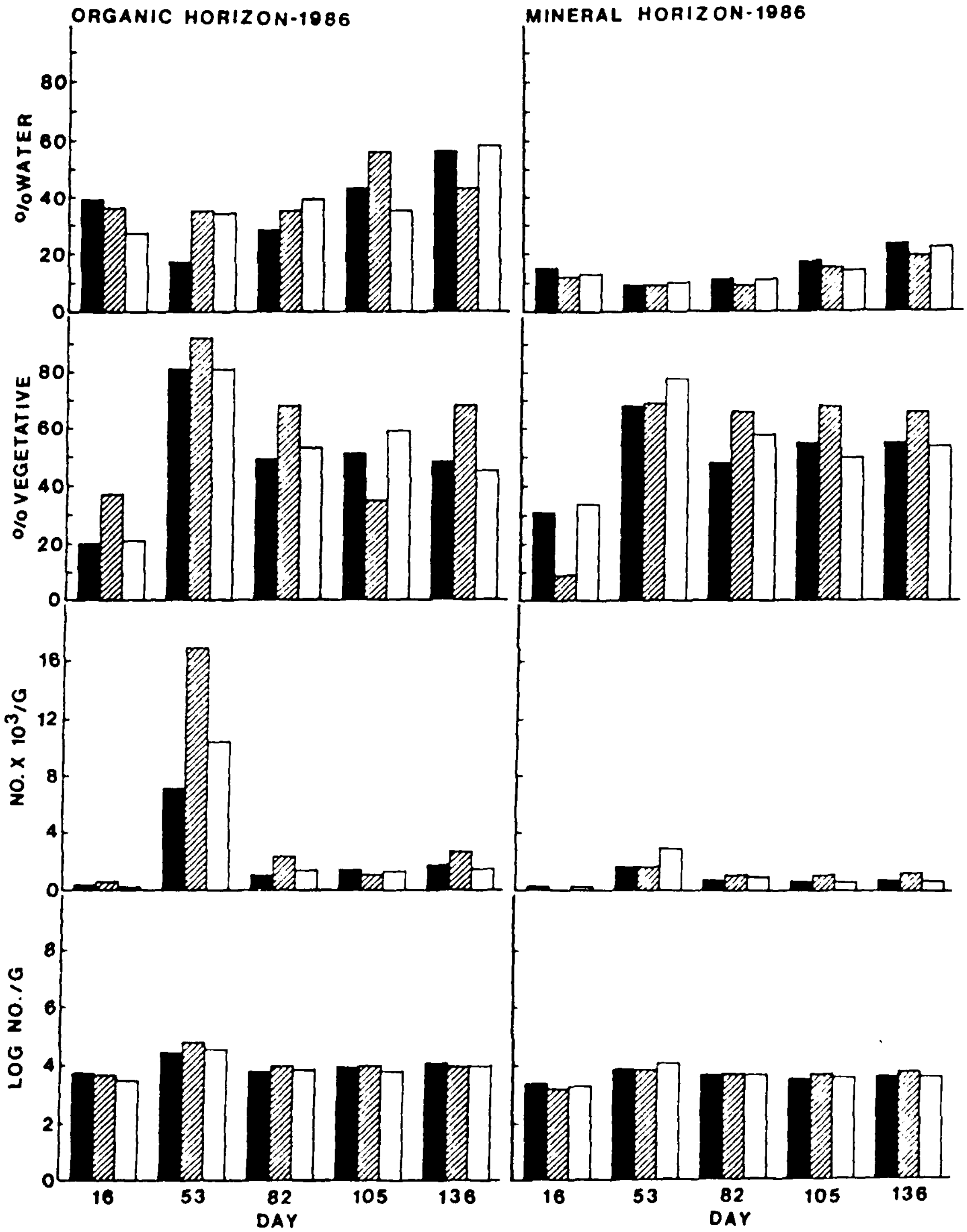
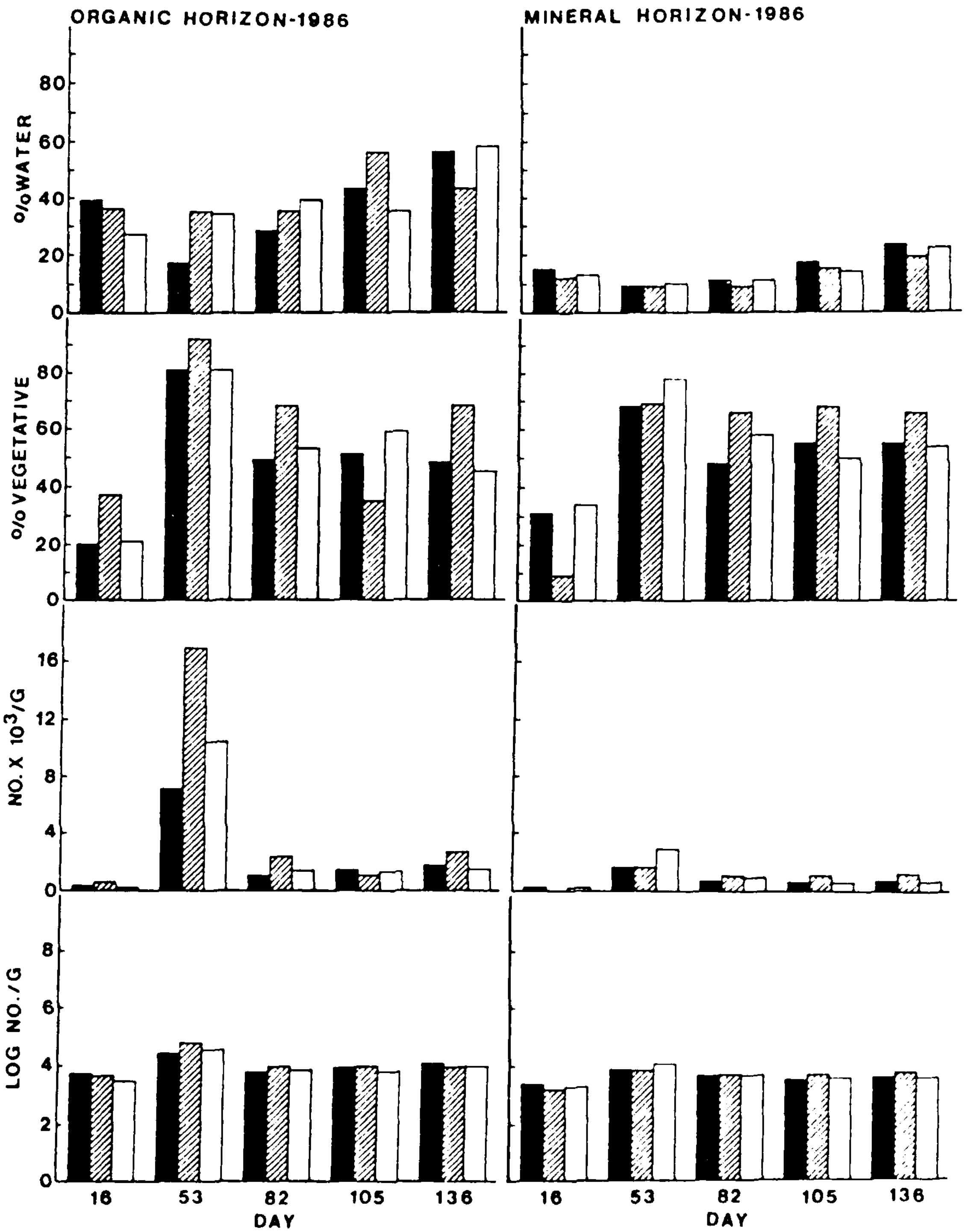


Figure 17. Summary of 1986 amoeba counts, given both as log counts and absolute numbers, % vegetative amoebae and % soil water. Note that the absolute number scale differs from previous years.



Appendix: IITRI provided the following procedures for testing growth in electric fields.

TEST SETUP

BAND

MATCHED E-FIELD PROTOCOL

1) Measure maximum E-field in soil using 1 meter probe - E.

2) Multiply E-field value by 0.15 to determine the minimum required drive voltage, V_{DR} (min).

$$V_{DR} \text{ (min)} = E \times 0.15 \text{ (volts)}$$

3) Locate collector electrodes in line with the maximum E-field in the earth, and spaced far enough apart to generate a voltage across a 2000 ohm resistor which is greater than or equal to V_{DR} (min). See Figure 1.

4) Measure and record electrode spacing and the open circuit (no load) electrode voltage, V_{OC}.

5) Connect the test cell and monitoring box to the electrodes. Refer to Figure 2. While monitoring the voltage across the test cell only, V_{CL}, adjust the variable resistor so that the cell voltage is equal to the value given by the following formula:

$$V_{CL} = E \times 0.113 \text{ (volts)}$$

6) With the cell voltage set, measure and record the voltage across the 100 ohm series resistor, V_R. This allows calculation of the cell current and current density.

7) Measure and record the electrode voltage with the test cell and monitoring box connected and adjusted as per Step 5, V_{DR}.

MATCHED CURRENT DENSITY PROTOCOL

1) Measure maximum E-field in soil using 1 meter probe - E.

2) Locate collector electrodes in line with maximum E-field with a separation of 1 meter.

3) Measure exact electrode spacing and open circuit (no load) electrode voltage, V_{OC}. Measured voltage should be within a few percent of that measured in Step 1. If not, correct electrode spacing as appropriate.

4) Connect current-limiting test ohamber (see Figure 3) to electrodes. Place the current limit select swtich to the 250 megohm position
2.5M

5) Measure and record the voltages across the test cell, V_{CL}, the resistor, V_R, and the electrodes V_{DR}, using the test point jacks. Refer to Figure 3 for test point numbering.

The voltages across the resistor and across the electrodes should be close in value to V_{OC} from Step 3.

$$V_R \simeq V_{DR} \simeq V_{OC}$$

The voltage across the test cell will be much lower, and can be estimated as:

$$V_{CL} \simeq 0.6 \times 10^{-3} \times V_C \text{ (volts)}$$

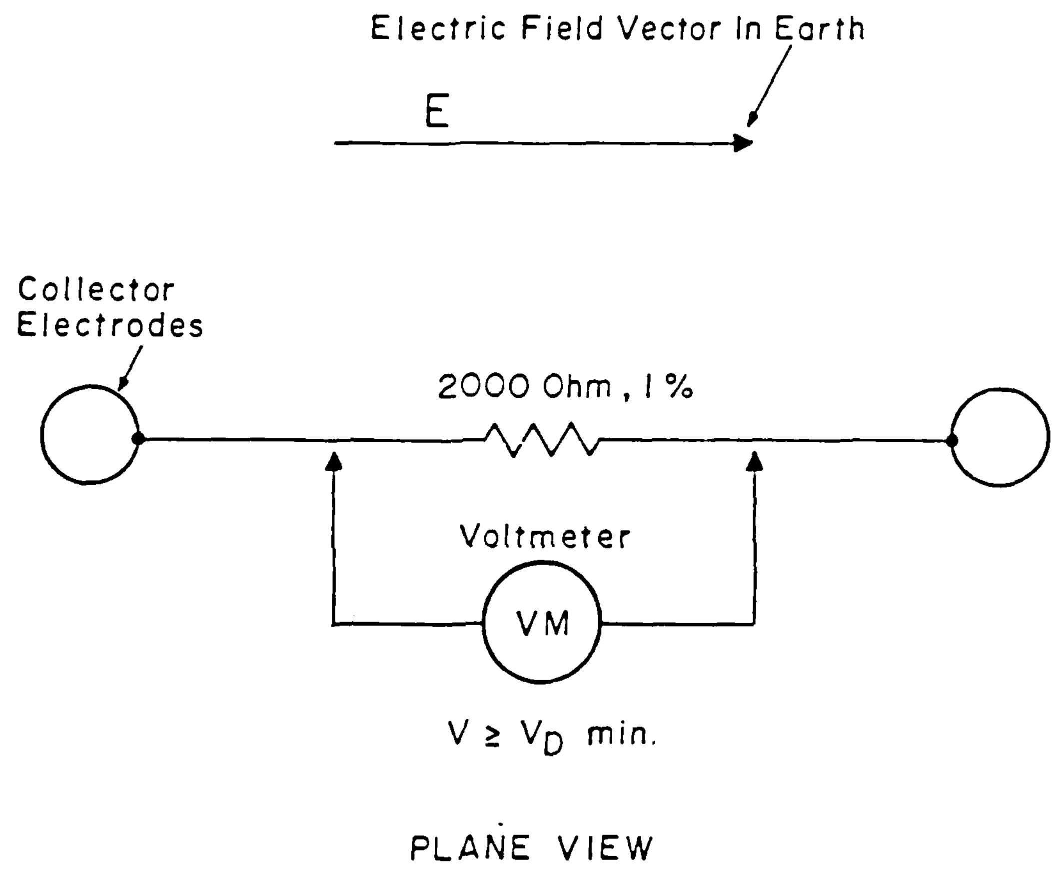
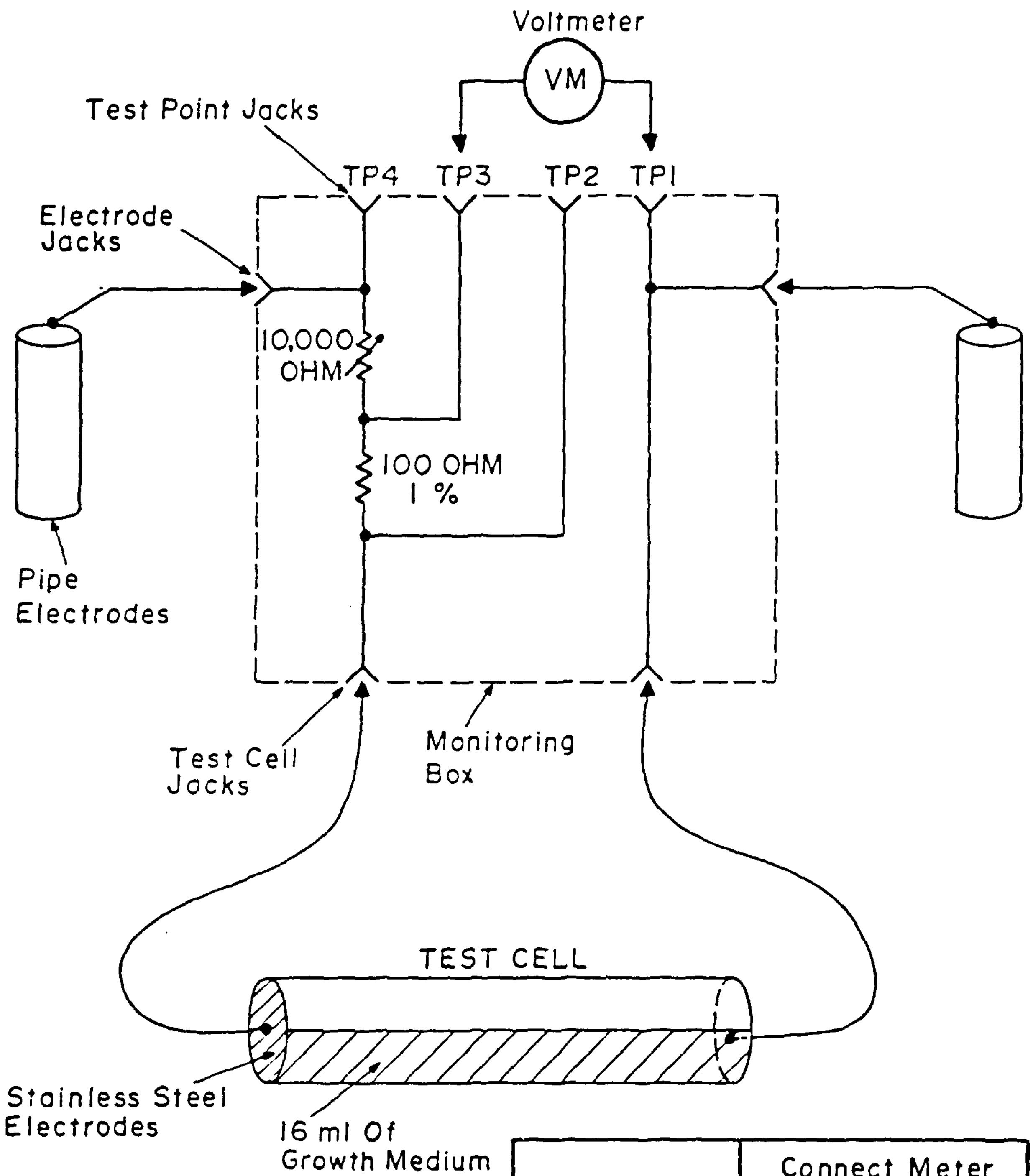
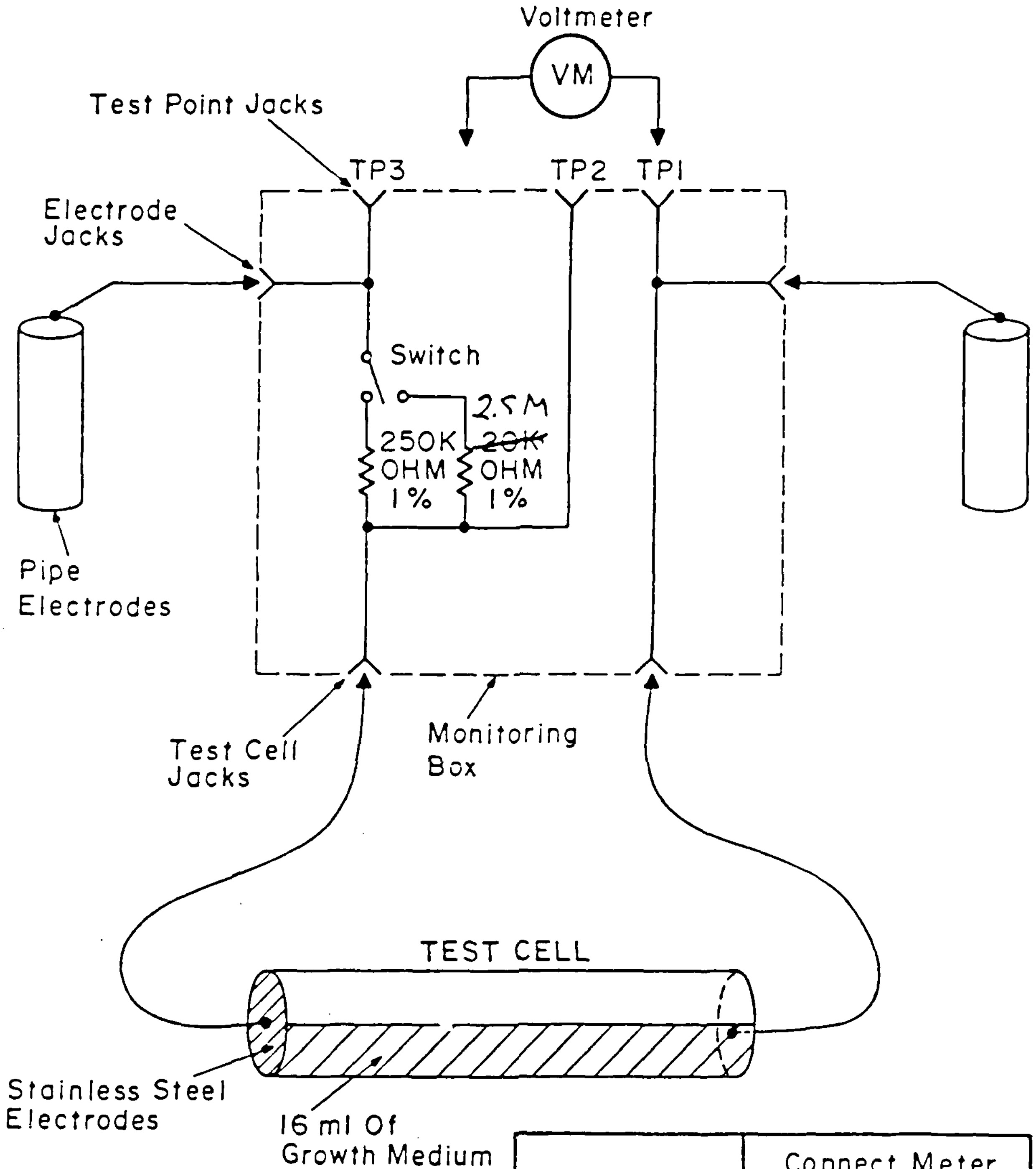


Fig. 1 DETERMINATION OF DRIVE VOLTAGE



To Measure	Connect Meter Across
V_{CL}	TP1 - TP2
V_R	TP2 - TP3
V_{DR}	TP1 - TP4

Fig. 2 TEST CELL HOOKUP FOR MATCHED E-FIELD PROTOCOL



To Measure	Connect Meter Across
V_{CL}	TP1 - TP2
V_R	TP2 - TP3
V_{DR}	TP1 - TP3

Fig. 3 TEST CELL HOOKUP FOR MATCHED CURRENT DENSITY PROTOCOL

Michigan State University

East Lansing, Michigan 48824

Subcontract No.

E06549-84-C-004

ELF Communications System Ecological Monitoring Program:

Soil and Litter Arthropoda and Earthworm Studies

Tasks 5.3. and 5.4.

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Principal Investigator

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TABLE OF CONTENTS

ABSTRACT

Test and Control sites were sampled, as in previous years, from early May to late October. Samples of arthropods and earthworms were obtained at intervals of two weeks. Surface-activity of arthropods was assessed by consecutive day and night pit-trap samples taken once a week. Based on two to three years of pre-ELF data (not all 1986 samples have been identified to date), considerable year-to-year variation in community structure and abundance of various species populations was shown to occur. Several species of Collembola, mites and earthworms exhibited either synchronous numerical fluctuations in Test and Control, or showed highly synchronized appearance and development of their life stages. Overall changes in community structure, as well as species population phenologies, will be useful as indicators of potential ecological changes in the future. Leaf litter inputs did not differ significantly either between sites or years. Litter decomposition studies were reduced in scope, but were also tightened to provide data sensitive to analysis of potential future changes.

SUMMARY

Sampling schedules in 1986 were adhered to rigorously in 1986, from May 7 to October 22 (2-week intervals for arthropod and lumbricid samples, weekly collection of diurnal and nocturnal pit-trap samples). Unlike previous years, a third B horizon sample (to a depth of 30 cm below the A horizon) was extraction of earthworms, in order to better quantify vertical distribution during drought.

Environmental variables (air and soil temperature, soil and litter moisture, RH, precipitation) were monitored throughout the season. In 1986, precipitation was severely deficient, particularly during the first half of the season. As a result, litter remained dry into July, and soil moisture dropped below 25% as early as May. Epigeic earthworms were essentially absent from leaf litter during this time, while endogeic species retreated, to species-specific degrees, into the A-30 cm stratum and probably even deeper.

Earthworm population cycles were shown to differ between years, timing and extent of cocoon production, and of juvenile emergence the following year, being governed mainly by moisture conditions. Dendrobaena octaedra (epigeic) occurs in both sites and will provide a means of comparing pre- and post-ELF population behavior. So will two dominant Aporrectodea spp. (A. tuberculata in Test and A. turgida in Control) which proved to be very similar in their biology and response to moisture stress.

Collembolan communities fluctuated from year to year in terms of overall densities (higher in 1985 than in 1984) and in terms of community structure and composition. While the most common species in both sites

remained dominant, a large number of rare species were first recorded from 1985 samples, and several species encountered in 1984 were no longer present in 1985 soil or litter samples. Among mite taxa which have been analyzed in detail, three species exhibited very similar life cycles in Test and Control, with synchronous emergence of larvae and maturation patterns in both sites.

Unlike 1984, pit-traps were provided with barriers in 1985. As a result, catches of surface-active arthropods were increased. Although statistical treatment of data is not yet complete, it is clear that diel activity patterns are species-specific, and may vary from year to year in response to day and night temperature regimes. Flexible species, both of Collembola and Carabidae, switch to diurnal activity following cold nights, while less plastic species respond with overall activity increases or decreases. Community structure of surface-active groups was also variable from year to year, which is only partly explained by effects of barriers. Rather, it is likely that such variability is due to normal yearly population surges and declines.

Litter inputs in 1986 were similar to those observed previously, and differed significantly neither between sites nor between years. Data on litter standing crops for 1986 are as yet incomplete. We implemented a litter-washing technique which will improve data accuracy and will allow a more realistic estimate of forest floor litter decay rates. However, not all samples could be processed prior to the onset of cold weather, and the remainder were stored until spring of 1987. We have discontinued litterbag studies, which yielded unrealistic decay rates, as well as nutrient analyses due to lack of manpower and insufficient control over data quality. Leaf-

pack data, however, indicated that this method will be worth using in the future. Both sun and shade leafpacks decayed at equal rates in Test and Control during the first year of decomposition on the forest floor.

I. REVIEW

The pre-ELF years have now stretched from late 1983 through 1986. Most of the faunal data through 1985 have now been finalized, and once 1986 are available, we will have three full seasons' worth of background data against which to measure potential future disturbance.

During the evolution of this project, we have kept to our original objective which dealt mainly with monitoring of arthropod and earthworm populations. Process-oriented work elements were phased in during earlier years, but had to be pared down in 1986 for several reasons. Nutrient analyses, for instance, had to be abandoned because of manpower shortage as well as lack of quality control over analytical results. However, we believe that the work elements which have been consistently pursued, particularly those which have been improved in terms of methods and validative procedures, justify the deletion of selected litter breakdown studies and the concurrent shift toward intensified documentation of invertebrate dynamics.

A slightly revised version of objectives (as listed in the last report), which in fact returns this project to its initially contracted goals, is given in Fig. 1. All objectives listed there now form the core of our investigation, and the extent and usefulness of the data base to the overall goals of Project ELF will be presented in the remainder of this report.

We realize the importance of publishing results in the open literature, and several papaers have so far been either published, accepted for publication, or have been submitted. We have hesitated to finalize certain major sets of data either because complex statistical analyses are still pending, or because yet another year of pre-ELF monitoring loomed ahead of us. We have recently decided that, once 1986 faunal data have been completed, we will

possess a publishable pre-ELF data base spanning three full years, which allows year-to-year comparison of all major forest floor biota we are monitoring.

SITES

TEST AND CONTROL

SITE COMPARISON
Vegetation and soil analysis: completed

ENVIRONMENTAL MONITORING

- -Precipitation
- -Air temperature and RH
- -Soil temperature
- -Soil moisture
- -Litter moisture

LUMBRICIDAE

- -Distribution
- -Community structure
- -Seasonal densities
- -Phenology of species

ARTHROPODA

- -Community structure
- -Population dynamics
- -Distribution
- -Diel activity patterns

DECOMPOSITION

- -Litter input
- -Natural litter turnover
- -Litter mass loss

Fig. 1. Ecological monitoring objectives in Test and Control sites.

II. ENVIRONMENTAL MONITORING

1. Temperature and humidity

Nineteen hundred and eighty-six was the first year in which virtually continuous records were obtained in both sites with Omnidata logging equipment. Temperature sensors that were to be installed at various depths in Test and Control were compared in the laboratory, showing that minor differences existed between them. Those used for air, soil surface and A horizon (5 cm depth) monitoring were comparatively the same (≤ 0.5°C difference between Test and Control). Sensors to be installed at 15 and 25 cm depths differed occasionally, but not consistently, by up to 1.5°C. We concluded that daily averages of readings taken at 2-hour intervals in both sites would yield valid temperature profiles over the season.

The voluminous data are not shown here, but for example daily means of RH and temperature for a randomly chosen period of 14 days are given in Table 1. Climatic variables were clearly similar in both sites. Relative humidity tended to be slightly higher in Control, but standard hygrothermograph charts showed that daily maxima and minima occurred simultaneously and had the same magnitudes in Test and Control. Air temperatures consistently differed by ≤ 0.5°C, and soil surface means by < 1.0°C. Soil temperatures at 5 and 15 cm depth tended to be higher in Test, which is not totally explained by sensor inconsistencies. Rather, the difference is likely to be real and due to the water-holding capacities of the A horizon (higher in Control, see Section II.3.).

In summary, air (confirmed by hygrothermograph reading) and soil surface temperatures under litter cover were reliably recorded throughout the 1986 season and are now also available for much of the 1985 season.

Table 1. Example of daily mean temperature and RH in Test and Control, June 1-14, 1986.

	% RH		°C Air		0 cm		5 cm		15 cm		25 cm	
	T	C	T	C	T	C	T	C	T	C	T	C
6/1	78	78	8.0	8.5	11.7	11.5	13.3	12.0	14.2	13.2	13.0	12.0
6/2	57	62	8.6	8.3	10.2	10.2	11.1	10.2	12.4	11.5	11.4	10.8
6/3	54	53	14.4	14.6	13.0	12.9	12.1	11.4	12.5	11.9	11.3	10.6
6/4	76	73	14.3	14.8	13.9	13.4	13.6	12.5	13.7	13.0	12.0	11.3
6/5	62	63	10.2	10.6	11.5	11.3	12.0	11.0	12.9	12.0	11.7	10.8
6/6	64	63	12.1	12.2	12.2	11.4	12.1	10.8	12.8	11.6	11.3	10.6
6/7	89	91	14.2	14.2	13.3	12.7	13.0	11.9	13.3	12.3	11.6	11.0
6/8	65	69	13.9	13.9	13.3	12.8	13.2	12.2	13.7	12.8	11.8	11.1
6/9	61	62	14.0	13.5	13.1	12.5	13.0	11.9	13.5	12.6	11.8	11.2
6/10	84	88	13.5	13.0	13.2	12.4	13.1	11.9	13.7	12.6	11.9	11.2
6/11	95	99	9.8	10.2	11.6	11.5	12.9	11.7	13.7	12.7	12.1	11.3
6/12	94	98	10.8	11.0	11.0	10.8	11.4	10.4	12.6	11.6	11.1	10.5
6/13	68	68	14.0	14.0	12.9	12.7	12.7	11.9	13.1	12.4	11.4	11.0
6/14	78	79	13.3	13.6	13.0	12.7	13.1	12.2	13.6	12.8	11.8	11.3

2. Precipitation

The 1985 schedule of reading rain gauges four times during two consecutive days per week was continued through 1986, so that rainfall events surrounding pit-trapping days could be recorded.

Nineteen hundred and eighty-six totals (May 1 to October 20) were 36.2 cm for Test, 38.2 cm for Control, significantly below previous years' totals (Table 2). This severe deficit occurred mainly during May, June and July (Fig. 2) when rainfall events were not only few but also of short duration and generally low intensity.

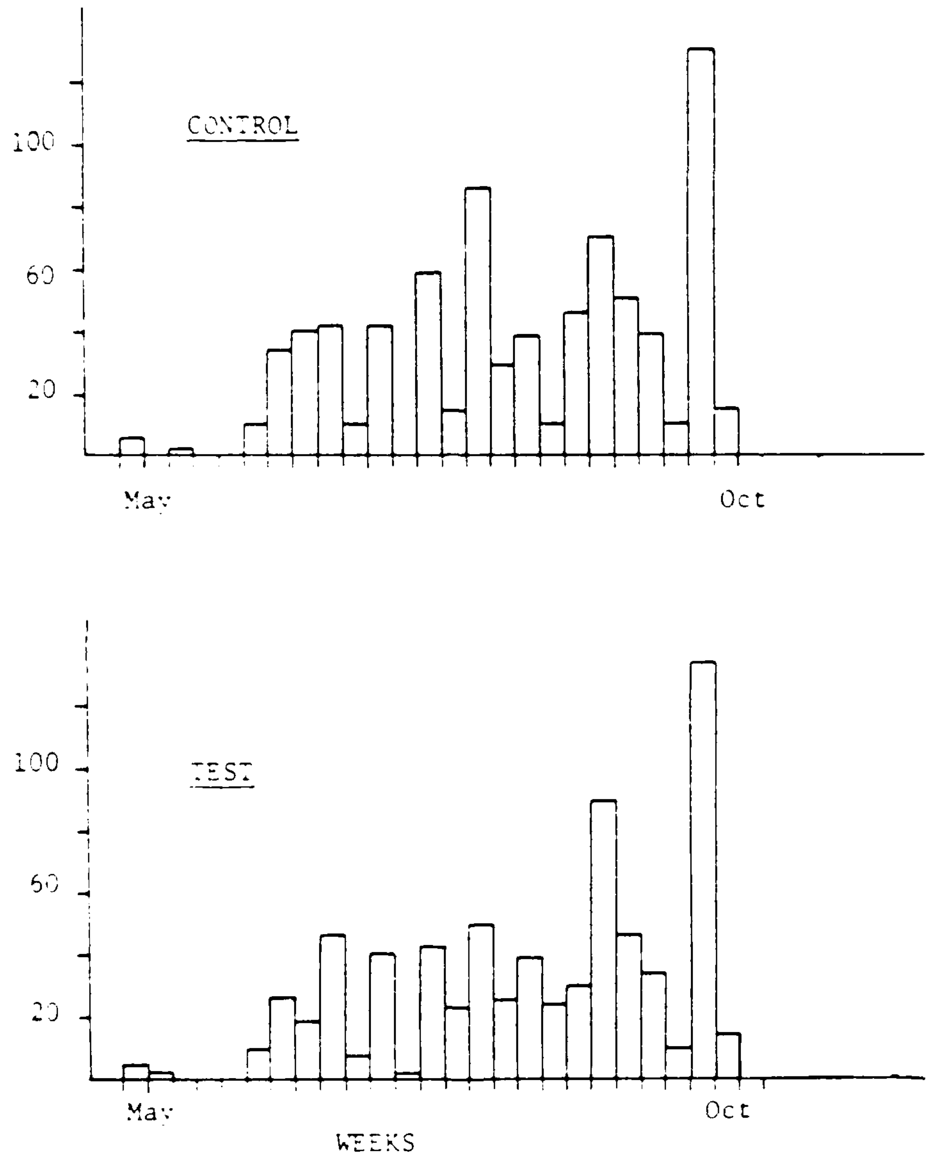

Fig. 2. Weekly precipitation totals, in mm, in 1986.

Table 2. Precipitation totals (in cm) for 1984-86, approx. from May 1 to October 15 of each year.

	1984	1985	1986
TEST	46.5	52.4	38.2
CONTROL	43.9	57.9	36.2

3. Litter and soil moisture

Snowmelt in mid-April of 1986 was rapid, and was followed by a severe drought in the study area. What rain did fall was mostly intercepted by the developing canopy. As a result, rain gauge data showed some precipitation in terms of weekly totals, but leaf litter remained dry until late July (Fig. 3) and was generally drier than on sampling days of previous year (Fig. 4).

Unlike 1984 and 1985 (Fig. 5), A horizon moisture fell below 25% at the beginning of the season in both sites and remained low in both sampled horizons through most of the summer (Fig. 6) due to subnormal precipitation. After 3 years of monitoring soil moisture, it seems clear that the Control A horizon possesses slightly greater water-holding capacities than that of the Test site (Figs. 5 and 6).

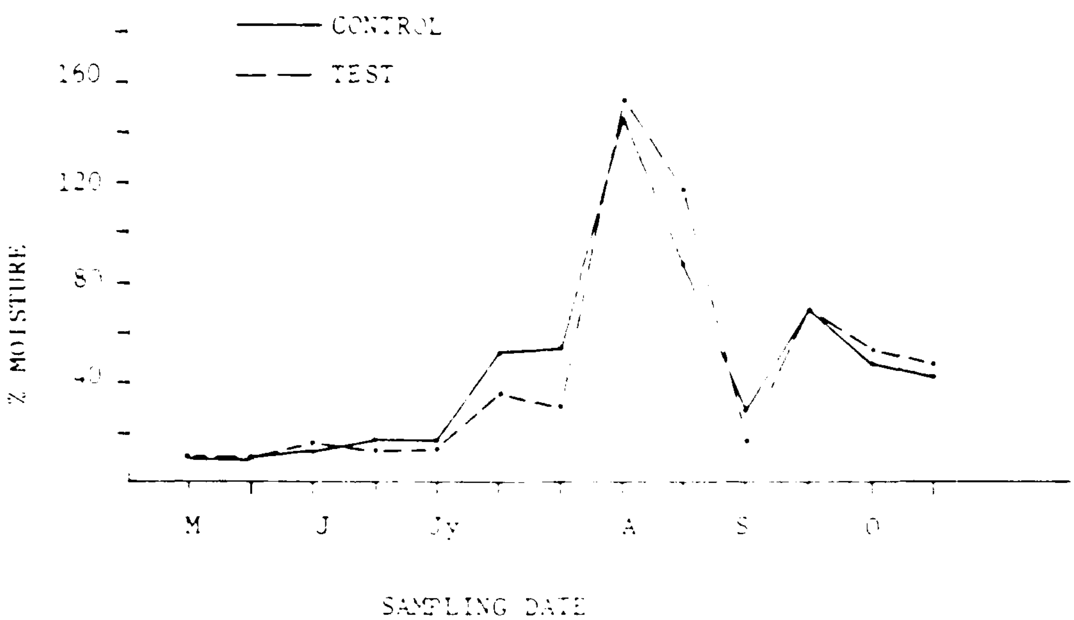

Fig. 3. Litter moisture, in % of dry weight, on faunal sampling dates in 198[illegible].

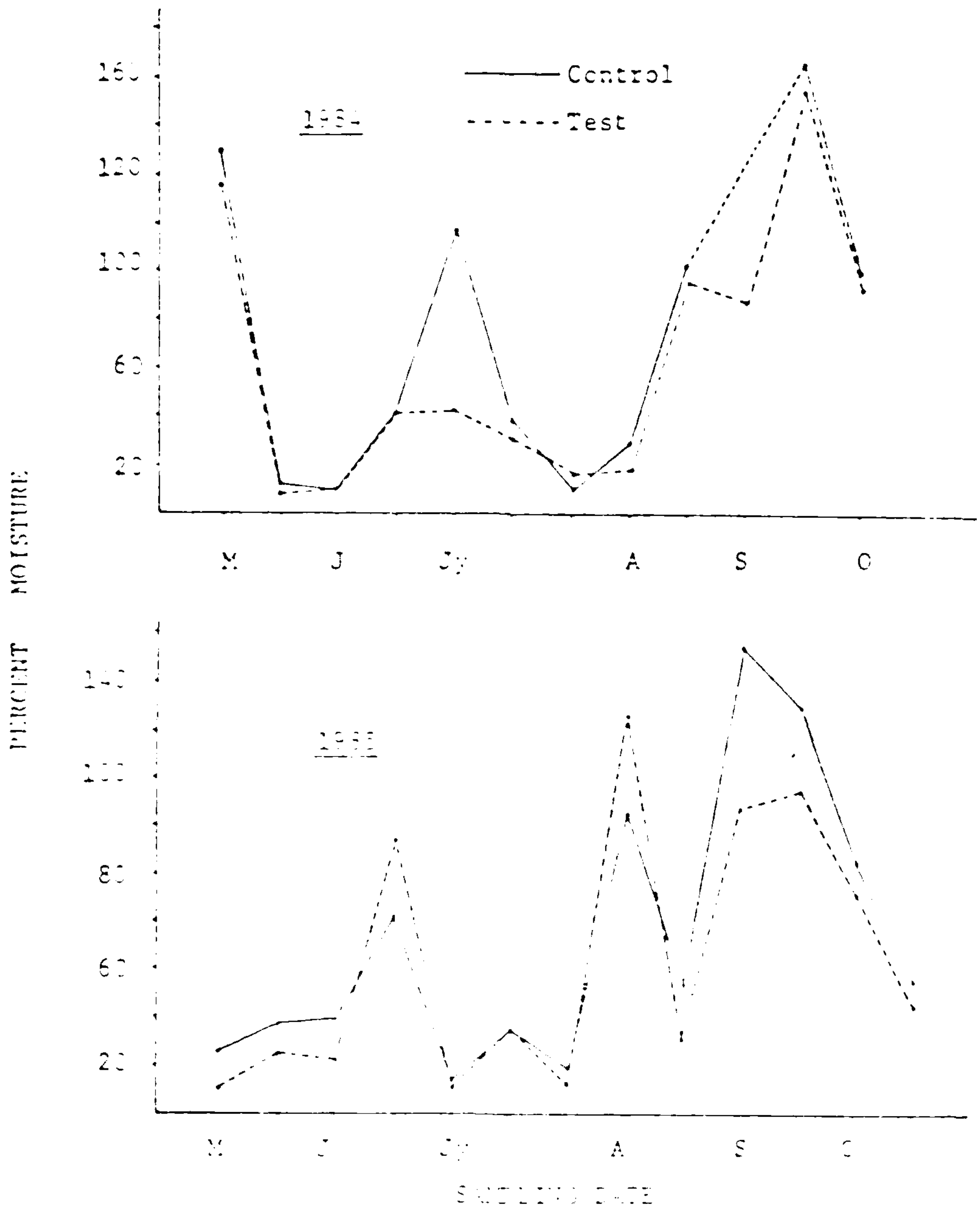

Fig. 4. Average litter moisture, in % of dry weight, in Test and Control on bi-weekly sampling dates in 198[illegible] and 19[illegible]. The Control peak in early July of 198[illegible] was due to a local rainfall event.

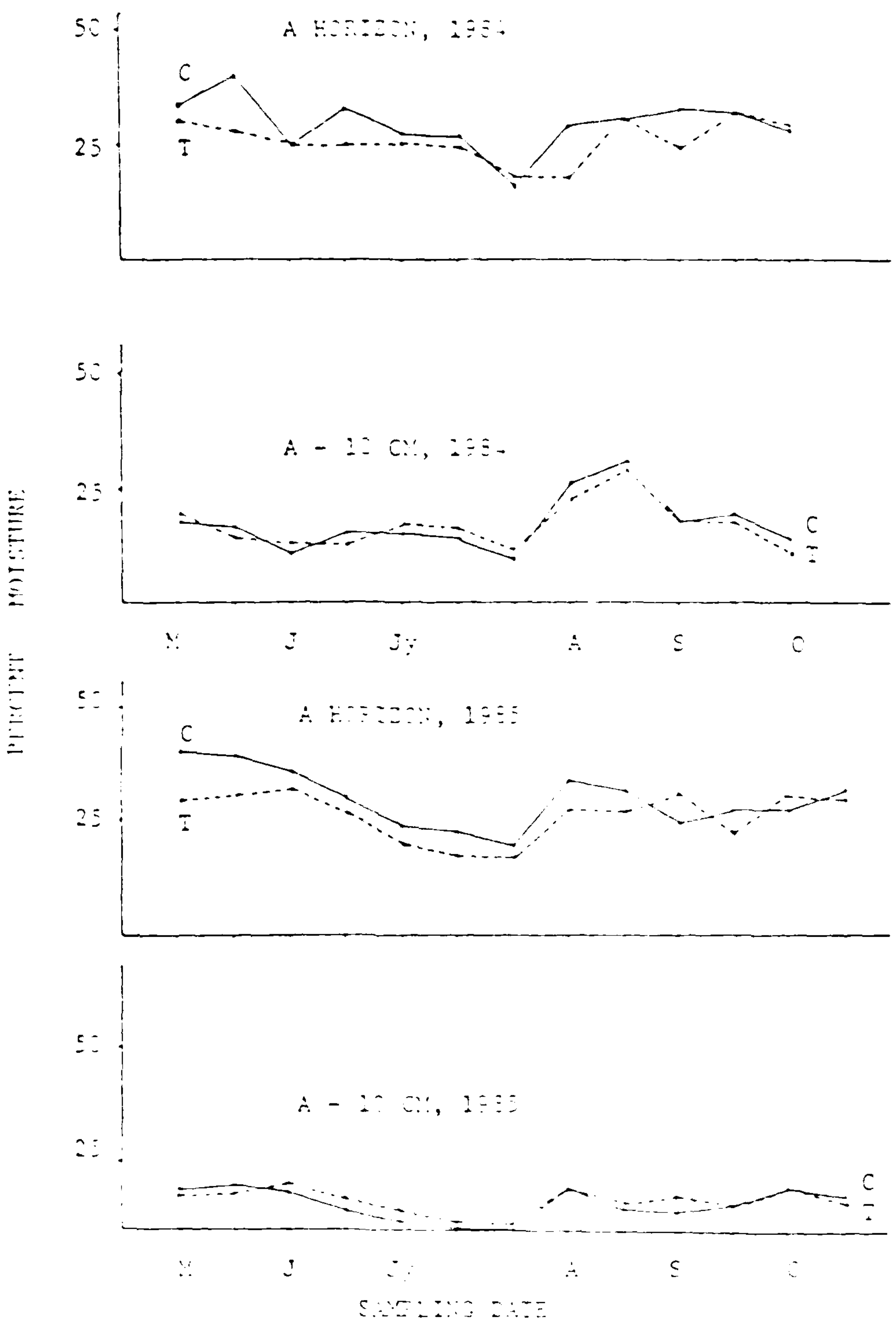

Fig. 5 . Moisture, in % of dry weight, of A and B horizons in 1984 and 1985.

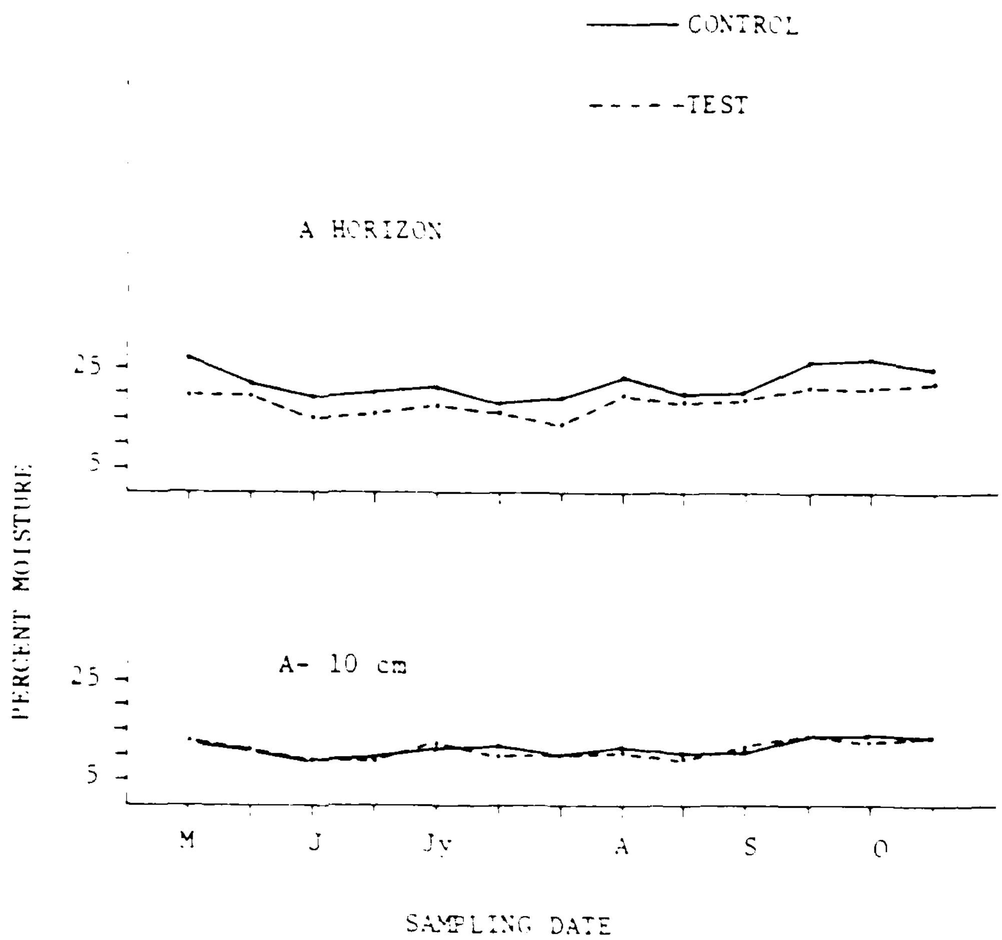

Fig. 5. Average moisture content, in % of dry weight, of soil in 1986.

III. SOIL AND LITTER ARTHROPODA

1. Extraction efficiencies

The variable extraction efficiencies for soil cores discussed in our last report prompted us to perform sugar-floatation after extraction of Test and Control samples on all 1986 sampling occasions. The method is very labor-intensive, because all specimens recovered after Tullgren-extraction have to be rehydrated on slide mounts. However, almost all can be identified to species, greatly adding to the accuracy of population estimates.

Although floatation samples have now been processed, the corresponding soil core (Tullgren) data are not yet available. For two dates, however, we give examples of efficiency values in Table 3. They confirm our earlier conclusions regarding the variability and degree of bias inherent in the Tullgren method, at least for the forest soils we deal with. Not only did efficiencies vary with date, they also varied greatly between individual samples. In agreement with Tamura (1976) and Loring (1979), Onychiuridae were affected most, particularly *Tullbergia* spp. which are the dominant members of the family in Test and Control soils.

Table 3. Percent efficiency of Tullgren extraction for Onychiuridae and Isotomidae, 1986 (N = 10 samples / site). E = total number Tullgren-extracted; F = total number recovered by floatation; % = percent of grand total successfully extracted in Tullgren funnels.

	ONYCHIURIDAE					ISOTOMIDAE				
	E	F	E+F	%	(Range, %)	E	F	E+F	%	(Range, %)
CONTROL										
June 16	280	424	704	39.8	(7.9-68.9)	74	7	81	91.4	(50-100)
July 28	112	492	604	18.5	(0-75.4)	24	11	35	68.6	(0-100)
TEST										
June 16	38	9	47	80.9	(0-100)	112	3	115	97.4	(0-100)
July 28	46	223	269	17.1	(0-64.7)	15	6	21	71.4	(20-100)

We do not have efficiency data for 1985 samples, and will discuss the Collembola of Test and Control with partial exclusion of Onychiuridae, as in our last report.

We have also made sample floatation after extraction a permanent work element of this project.

2. Status of data

All 1985 litter and soil samples have been sorted and identified to various taxonomic levels appropriate to the groups of interest. Complete identifications and summaries are available for Collembola. As proposed in 1985, focusing only on selected taxa among the diverse Test and Control Acari has allowed us to close earlier gaps considerably. "Species A" (Mesostigmata), Asca aphidioides and Nanorchestes spp.[1] have been processed, including 1985 material, which is made available in the present report. In the case of Rhagidiidae and Eupodidae, we have not yet had the benefit of the outside expertise we solicited, but are proceeding with identification of 1985 material.

Among samples taken in 1986, pit-traps were given priority as usual, and species determination of trapped material has begun. Rough-sorting of soil and litter samples from 1986 has just been initiated, and to save time identification of specimens will proceed simultaneously. Sugar-floatation samples from 1986 have long been sorted, and most of the specimens have been mounted and identified.

Overall, although the gap between sampling and data availability cannot be significantly reduced, we are accommodating additional work in terms of sorting and identification of floatation samples and of the more numerous pit-trapped specimens obtained by barrier-trapping (begun in 1985).

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[1]The help of Dr. Lindquist in identifying and characterizing Nanorchestes spp. is here gratefully acknowledged.

3. Collembola communities of Test and Control

i. Density and dominance

The two full-season sampling periods of 1984 and 1985 allow a first assessment of year-to-year fluctuations in Test and Control communities. Table 4 details some of these changes. In terms of species composition, 15 species were added to the list for Test, while 3 were no longer recorded in soil or litter samples. In Control, 20 species were obtained for the first time in 1985, while 8 were no longer recorded. In most cases, they represent rare members of the communities. Notable exceptions are Folsomia bisetosa and Neelus minimus in Test (from zero to > 3% overall dominance value in 1985); and Anurophorus altus and, again, Neelus minimus in Control (Table 4).

Overall, I. notabilis was the single most common species in both sites and both years. Its dominance value in Control increased, together with that of T. flavescens and F. bisetosa. In both sites, the majority of species showed some change in relative dominance (Table 4), a direct reflection of year-to-year fluctuations in abundance.

Again excluding Onychiuridae, dominance relationships at the family level did not vary drastically between years (Table 5). It should be noted that all dominance increases and decreases in Test corresponded to increases and decreases in Control, often of similar magnitude. Furthermore, total numbers in 1985 were higher than in 1984 in both sites (by 43% in Test, by 38% in Control).

Overall density increases could be attributed mainly to the dominants in both sites, although significant contributions were also made by hypogastrurids, neelids and isotomids not so far encountered (Table 6). Folsomia bisetosa and Neelus minimus provide examples for Test, W. intermedia, A. furcifera and N. minimus for Control.

ii. Diversity and similarity

Year-to year fluctuation in species composition and relative abundance affected the diversity measures of these communities (Table 7). The leaf litter subcommunities were less diverse in 1985 than in 1984, dominance indices correspondingly increasing in both Test and Control. The single most likely cause for these changes was the increased abundance of *I. notabilis* (more than 2-fold in Test, 3-fold in Control, Table 6). Similarity indices for these subcommunities were not greatly altered.

Table 4. List of Collembola species and percent dominance (D% within family in parentheses) in Test and Control, 1984-85. 0 = absent; - = D< 1%.

	TEST		CONTROL	
	1984	1985	1984	1985
SMINTHURIDAE				
Sminthurinus henshawi (Folsom)	5.2(57.0)	6.2(54.8)	5.1(75.2)	5.9 (60.8)
S. macgillivrayi (Banks)	- (2.9)	- (5.9	- (0.4)	- (0.3)
S. quadrimaculatus (Ryder)	0 0	0 0	- (0.1)	- 0
Sminthurides lepus Mills	- (3.2)	- (3.8)	- (0.4)	- (1.7)
Dicyrtoma aurata (Mills)	0 0	0 0	- (6.3)	- (2.5)
D. marmorata (Packard)	0 0	0 0	0 0	- -
Arrhopalites spp.	3.4(36.9)	3.6(35.5)	1.2(17.5)	3.0 (34.7)
Bourletiella hortensis (Fitch)	0 0	0 0	- (0.1)	0 0
B. russata Maynard	0 0	0 0	- (0.1)	0 0
ISOTOMIDAE				
Isotoma notabilis Schaeffer	39.1(74.3)	40.4(74.5)	44.4(59.7)	58.0 (74.7)
I. nigrifrons Folsom	3.1(5.8)	2.2(4.0)	1.5(2.0)	- (5.4)
I. viridis Bourlet	- (0.3)	- (0.8)	0 0	- -
I. pseudocinerea (Fjellberg)	0 0	- (0.2)	0 0	0 0
Folsomia nivalis (Packard)	- (0.6)	2.2(4.0)	6.1(8.2)	3.8 (4.9)
F. bisetosa Gisin	0 0	3.9(7.1)	- (0.2)	5.1 (6.6)
Anurophorus altus Chris. & Bell.	0 0	0 0	0 0	3.7 (4.7)
A. binoculatus (Kneseman)	1.0(1.9)	1.0(1.8)	2.1(2.8)	1.7 (2.2)
A. septentrionalis Palissa	0 0	0 0	8.6(11.5)	0 0
Isotomiella minor (Schaeffer)	8.3(15.7)	3.8(7.0)	10.2(13.8)	2.8 (3.6)
Cryptopygus exilis (Gisin)	0 0	- (0.2	0 0	0 (0.4)
C. decemoculatus (Folsom)	- (0.3)	0 0	0 0	- 0
Proisotoma minima (Absolon)	- (1.0)	- (0.5)	1.3(1.8)	1.8 (2.3)
ENTOMOBRYIDAE				
Tomocerus flavescens Tullberg	7.5(26.6)	10.6(41.4)	- (15.5)	1.0 (26.8)
T. lamelliferus Mills	3.1(10.8)	1.0(4.2)	- (7.3)	- (0.6)
Orchesella hexfasciata Harvey	3.7(13.1)	4.7(18.3)	1.4(22.7)	- (20.5)
Entomobrya comparata Folsom	- (2.3)	- (3.4)	2.2(36.5)	1.6 (43.3)
E. nivalis (L.)	- (2.8)	1.0(3.7)	- (2.2)	- (2.3)
E. purpurascens Packard	- (0.5)	- (0.1)	- (0.7)	- (0.3)
Lepidocyrtus violaceous Fourcroy	- (0.5)	- (1.1)	- (2.0)	0 0
L. helenae Snider	- (0.3)	- (0.5)	- (5.0)	- (4.0)
L. lignorum (Fabricius)	- (0.1)	- -	0 0	0 0
Pseudosinella violenta (Folsom)	12.1(43.0)	6.7(26.1)	0 0	0 0
Willowsia buski (Lubbock)	0 0	- (1.1)	0 0	- (2.3)
HYPOGASTRURIDAE				
Pseudachorutes saxatilis Macnamara	- (30.7)	0 0	1.4(20.1)	- (1.2)
P. indiana Christiansen & Bellinger	0 0	- (0.1)	- (0.1)	0 0
P. aureofasciatus (Harvey)	0 0	- (1.8)	0 0	- (1.2)
Neanura muscorum (Templeton)	- (42.6)	- (14.6)	- (9.2)	2.1 (27.7)
Xenylla acauda Gisin	0 0	0 0	2.3(33.6)	- -
X. pallescens (Scott)	0 0	- (0.7)	0 0	- (2.1)
Willemia intermedia Mills	0 0	1.4(27.1)	0 0	1.7 (22.1)
W. similis Mills	0 0	1.8(35.9)	0 0	- (0.1)
Anurida granaria (Nicolet)	- (13.2)	0 0	2.3(33.2)	0 0
A. pygmaea (Borner)	-	- (1.9)	- (3.3)	- (2.1)

Table 4. cont'd.

	TEST				CONTROL			
	1984		1985		1984		1985	
Anurida furcifera (Mills)	0	0	-	(18.0)	0	0	2.2	(28.8)
Friesea sublimis Macnamara	0	0	-	(0.1)	0	0	-	(6.5)
Paranura caeca Folsom	0	0	0	0	0	0	-	(2.1)
Odontella substriata Wray	0	0	0	0	0	0	-	(6.0)
NEELIDAE								
Neelus tristani	8.8	(99.9)	-	(2.8)	5.7	(92.5)	-	(0.1)
N. minutus (Folsom)	-	(0.1)	-	(5.0)	-	(7.5)	-	(2.2)
N. minimus (Willem)	0	0	3.5	(92.1)	0	0	2.7	(77.2)
N. snideri (Bernard)	0	0	-	(0.1)	0	0	-	(20.5)
ONYCHIURIDAE								
Tullbergia mala Christ. & Bell.	-	(37.8)	-	(44.7)	-	(74.2)	-	(68.0)
T. granulata Mills	-	(46.7)	-	(33.7)	-	(18.0)	-	(22.6)
T. yosii Rusek	-	(10.1)	-	(2.6)	-	(2.5)	-	(0.7)
T. clavata Mills	-	(3.7)	-	(6.4)	-	(2.5)	-	(3.2)
T. falca Christiansen & Bellinger	-	0	-	(2.1)	-	0	-	-
T. iowensis Mills	-	0	-	(9.4)	-	0	-	(3.0)
T. hades Christiansen & Bellinger	-	0	-	(0.1)	-	0	-	-
Onychiurus similis Folsom	-	(1.8)	-	(0.9)	-	(2.4)	-	(1.7)
O. encarpatus Denis	-	0	-	0	-	(0.4)	-	0
O. affinis Agren	-	0	-	-	-	0	-	(0.4)
O. parvicornis Mills	-	0	-	-	-	0	-	(0.2)

NOTE: Onychiuridae only used for family-level calculations.

Table 5. Annual average density (May - October) of collembolan families, 1984-85 (in parentheses, dominance in % of total N). Onychiuridae excluded.

	ANNUAL MEAN N / M^2 (D%)			
	TEST		CONTROL	
	1984	1985	1984	1985
Sminthuridae	268 (9.2)	472 (11.3)	247 (6.7)	496 (9.8)
Entomobryidae	821 (28.1)	1065 (25.5)	219 (6.0)	139 (3.7)
Isotomidae	1536 (52.7)	2261 (54.2)	2727 (74.3)	3937 (77.7)
Hypogastruridae	34 (1.2)	214 (5.1)	255 (7.0)	387 (7.6)
Neelidae	258 (8.8)	157 (3.9)	224 (6.1)	178 (3.5)
TOTALS	2917	4169	3672	5065

Table 6. Mean annual density/m^2 (May-October) of Collembola with $D < 1\%$ (see Table 4). 0 = absent; - = $< 1/m^2$.

	LITTER				SOIL			
	TEST		CONTROL		TEST		CONTROL	
	1984	1985	1984	1985	1984	1985	1984	1985
S. henshawi	36	39	57	90	117	219	129	212
Arrhopalites spp.	20	10	2	4	79	158	42	46
I. notabilis	120	284	291	879	1021	1400	1338	2062
I. nigrifrons	10	25	13	6	79	65	42	15
I. nivalis	-	7	15	21	8	89	208	173
F. bisetosa	0	-	0	9	0	162	-	250
A. altus	0	0	0	52	0	0	0	135
A. binoculatus	9	2	40	27	21	39	37	62
A. septentrionalis	0	0	27	0	0	0	288	0
I. minor	0	-	-	-	242	158	375	142
P. minima	3	3	3	9	13	8	46	81
T. flavescens	56	83	17	16	163	358	17	35
T. lamelliferus	14	14	8	-	75	31	8	0
O. hexfasciata	16	22	8	8	92	173	42	31
E. comparata	19	21	71	51	0	15	8	31
P. violenta	3	-	0	0	350	278	0	0
P. saxatilis	2	0	10	-	8	0	42	4
N. muscorum	-	4	-	7	-	27	-	100
X. acauda	-	0	77	-	4	0	75	0
W. intermedia	0	-	0	-	0	58	0	85
W. similis	0	0	0	-	0	77	0	0
A. pranaria	-	0	-	0	4	0	83	0
A. furcifera	0	0	0	-	0	39	0	112
N. tristani	7	-	8	-	-	4	-	0
N. minimus	-	3	-	18	0	142	0	119
T. mala	-		-		1080	1708	5850	4381
T. granulata	-		-		1329	1285	1421	1458
T. yosii	-		-		275	100	195	39
T. clavata	-		-		104	246	196	204
T. iowensis					0	358	0	192

The soil subcommunity showed an opposite trend, with slight increases in diversity and reduced dominance (Table 7), due to the contribution of several species first recorded in 1985, and to the less pronounced density increases of the dominant I. notabilis (Table 6). Again, similarity indices of the soil subcommunities of Test and Control remained overall at the same level as in 1984 (Table 7).

Using combined soil and litter abundance estimates for each species, these changes in community structure became attenuated, and Sorensen's as well as Bray-Curtis similarity indices (Table 8) showed little variation between years. Combined (litter + soil) measures of community structure represent an "average" of subcommunity characteristics; they seem a valid descriptive tool in view of the fact that many species, particularly I. notabilis, frequent both strata of the forest floor.

iii. Distribution of litter Collembola

Litter mass of randomly taken samples varies within each date, and exhibits seasonal fluctuations as well. A preliminary analysis of factors affecting collembolan distribution aimed to assess a potential correlation between sample litter mass and numbers of animals extracted from them. Regression coefficients for each date were first computed using total N Collembola and dry litter mass/-sample; without giving details, Table 9 illustrates results obtained for Test data (1985). On 4 dates, a significant positive correlation between litter mass and collembolan populations existed. However, more complex analyses will be needed to determine the effect of moisture (which may act as an equalizer), and to test these relationships for each family (Entomobryidae and Isotomidae show discrepant seasonal distribution). In addition, these analyses may be applied to single species populations, to help explain seasonal distribution and numerical fluctuation.

Table 7. Diversity and similarity indices of collembolan communities in Test and Control, based on total N/species. Onychiuridae excluded.

		LITTER				SOIL			
		TEST		CONTROL		TEST		CONTROL	
		1984	1985	1984	1985	1984	1985	1984	1985
Shannon-Wiener H'		1.02	0.76	0.85	0.63	0.88	1.03	0.87	0.91
Simpson dominance		0.16	0.30	0.27	0.52	0.21	0.13	0.23	0.30
Simpson-Yule div.		6.13	3.33	3.73	1.92	4.88	5.56	4.42	3.33
Bray-Curtis simil.	1984	0.53				0.64			
	1985	0.45				0.66			
Sørensen's simil.	1984	0.81				0.74			
	1985	0.79				0.77			

Table 8. Diversity and similarity indices based on combined litter + soil densities/ m^2 of each species in Test and Control. Onychiuridae excluded.

		TEST		CONTROL	
		1984	1985	1984	1985
Shannon-Wiener H'		0.93	1.04	0.94	0.91
Simpson dominance		0.20	0.19	0.23	0.35
Simpson-Yule div.		5.0	5.21	4.35	2.85
Bray-Curtis simil.	1984	0.63			
	1985	0.65			
Sorensen's simil.	1984	0.84			
	1985	0.85			

Table 9. Regression coefficients for N Collembola on litter mass (g dry); average % moisture obtained from 20 "litter moisture samples" on each date.

Date	mean % H_2O	range litter mass	range N Collembola	r
5/7/85	9	8-31	0-29	0.88*
5/20/85	25	11-32	-61	0.48
6/3/85	23	8-39	4-36	0.53
6/17/85	115	5-29	3-141	0.62
7/1/85	10	5-34	0-55	0.84*
7/15/85	35	10-32	19-177	0.57
7/29/85	11	10-32	0-24	0.29
8/12/85	167	1-34	14-87	0.87*
8/26/85	28	3-19	4-83	0.66
9/10/85	128	4-25	5-147	0.69
9/24/85	135	3-23	6-100	0.80
10/6/85	91	8-32	0-83	0.86*
10/20/85	55	14-40	3-139	0.73

*)significant at $P < 0.01$

iv. Seasonal abundance of common species

Isotoma notabilis, dominant in both sites, showed relatively snychronous density fluctuations in Test and Control leaf litter in 1985, while populations in soil fluctuated both more widely and more discrepantly (Fig. 7). The greater abundance of the species in 1985 (Table 6) is clearly shown in year-to-year comparison within each site. In Control (Fig. 8), the litter subpopulation tended to increase in July and August, while numbers in soil decreased. In late October (sampled only in 1985), the beginning of a winter movement into soil was indicated. In Test (Fig. 9), inverse abundances of litter vs. soil subpopulations were particularly clear in summer and fall of 1985.

Populations in leaf litter of both sites offer the most intriguing fluctuations, drastic decreases coinciding in July 1 and July 29 samples. No obvious correlation with litter moisture (Fig. 4) seems to exist in either 1984 or 1985. Temperature records show that the first and last samplings in July of 1985 were

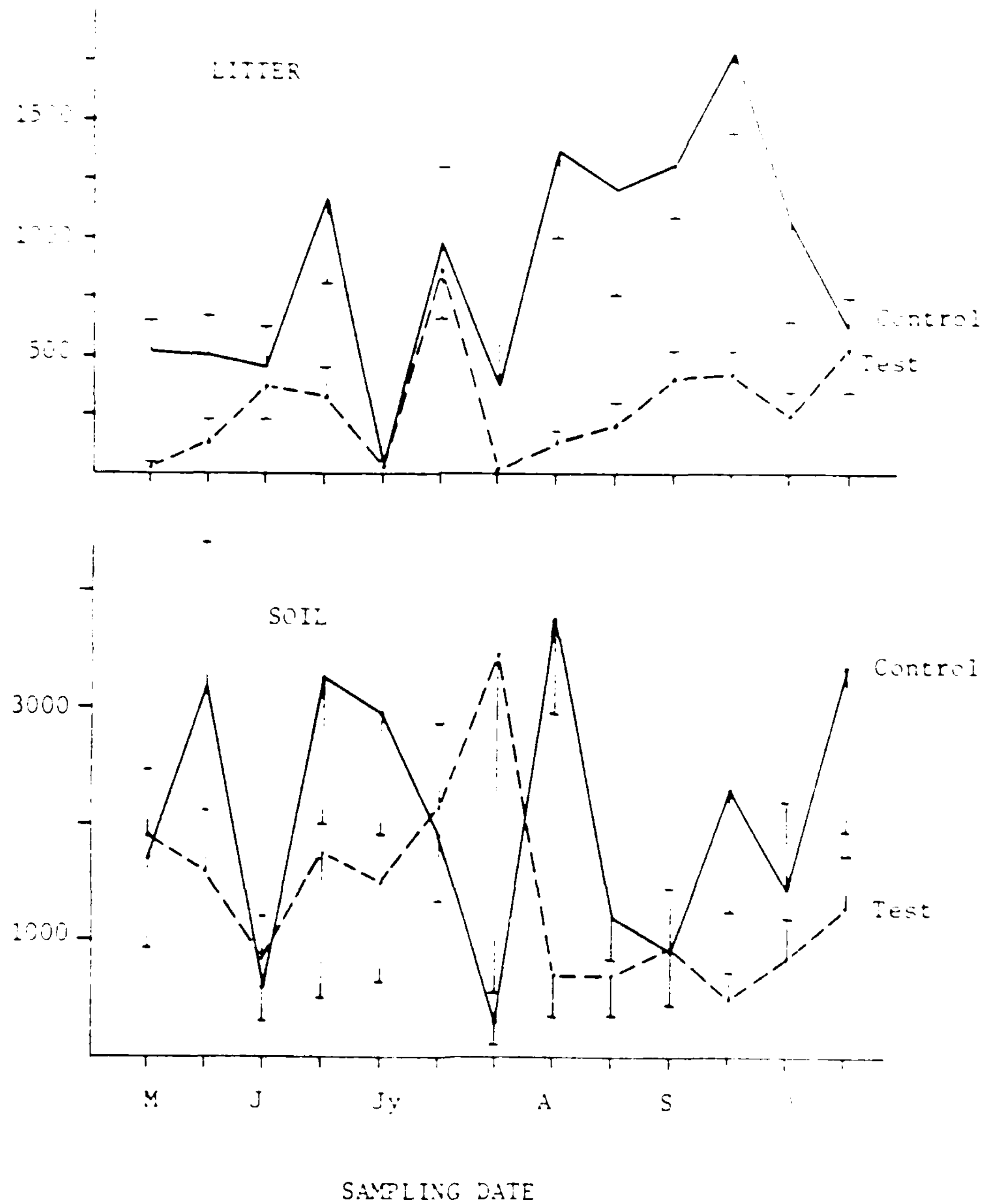

Fig. 7. Average density/ $m^2 \pm$ SE, of Isotoma notabilis in litter and soil, 1985.

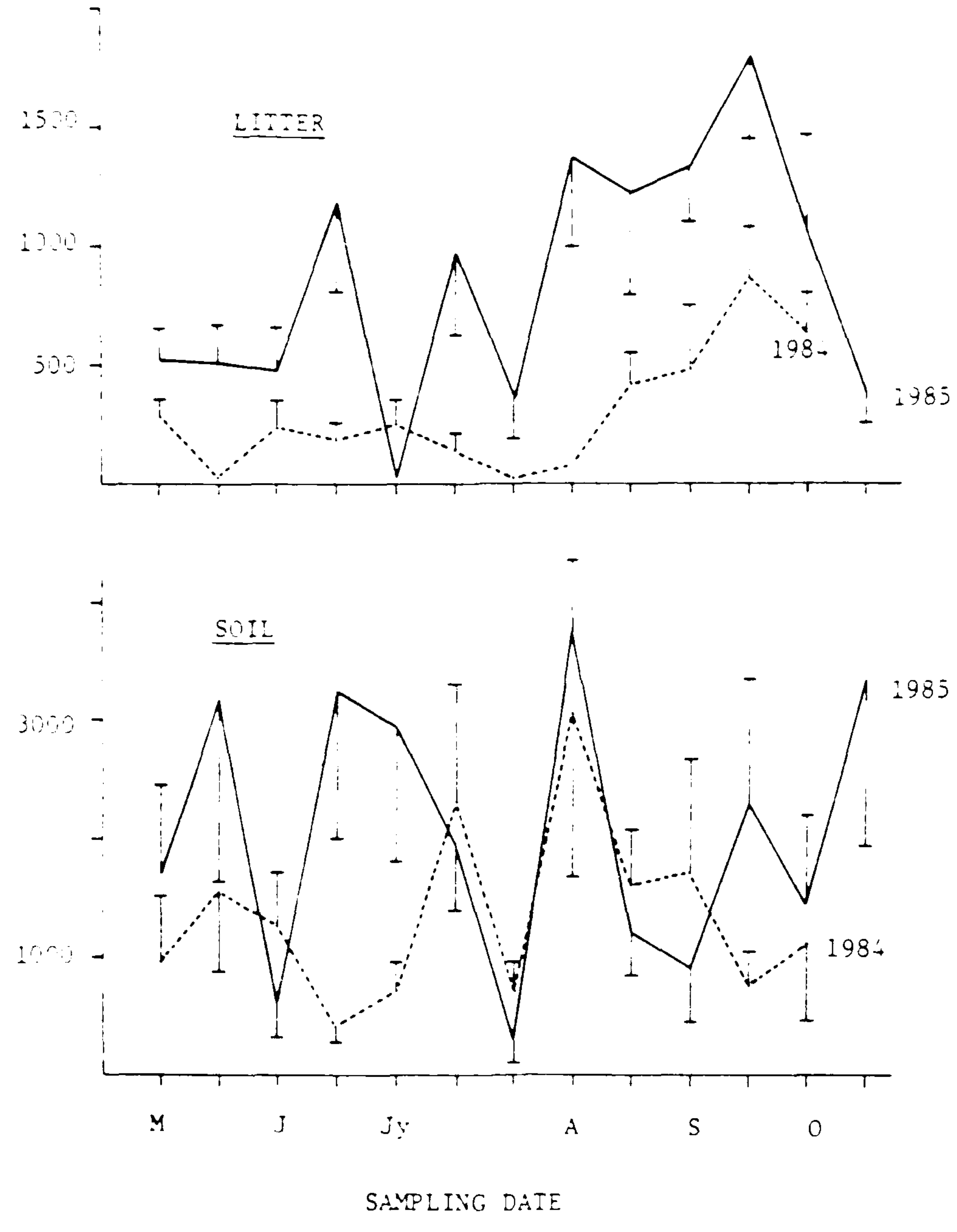

Fig. 8. Average densities / m^2 $\pm$ SE of Isotoma notabilis in Control litter and soil, 1984 and 1985.

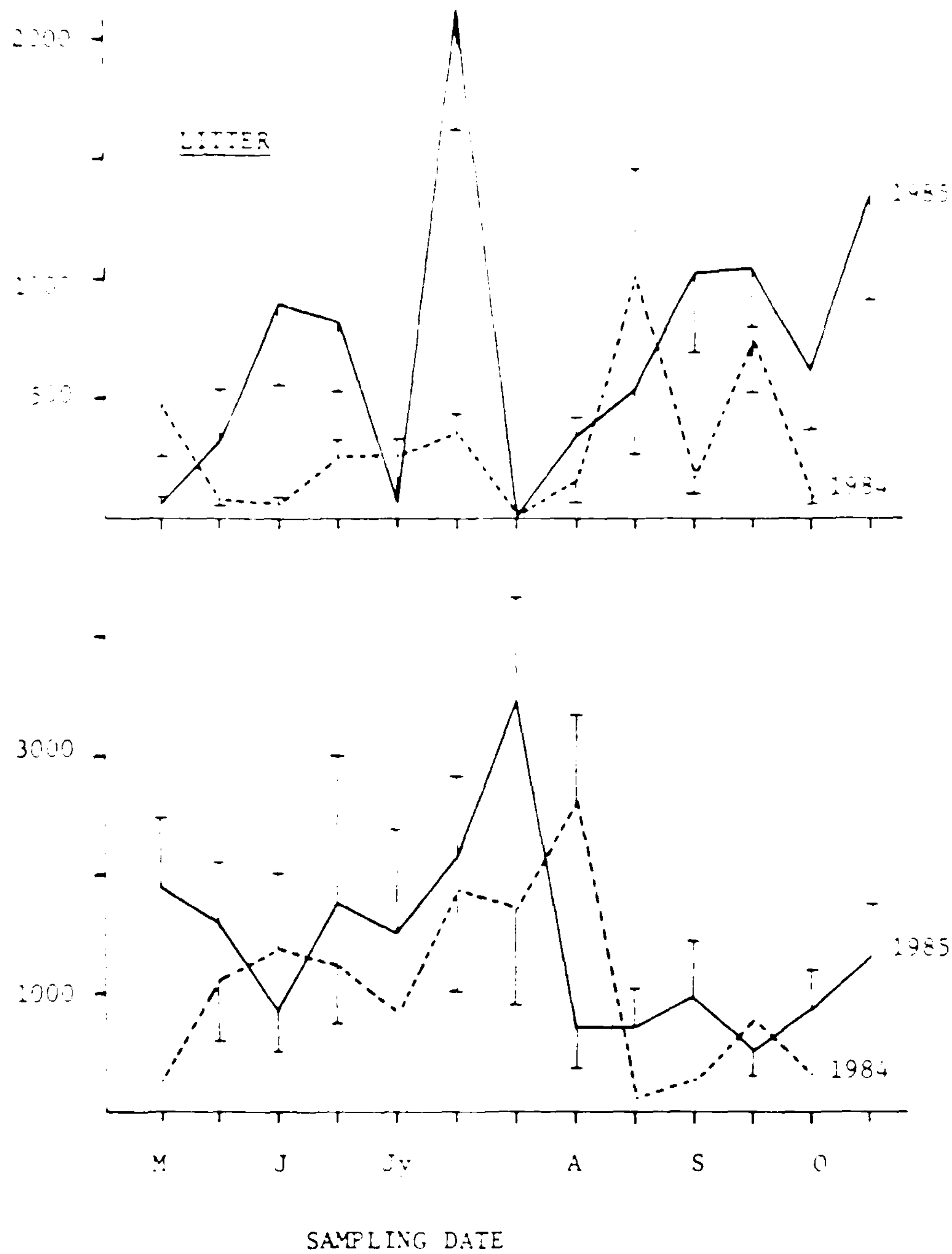

Fig. 9. Average densities / m^2 ± SE of Isotoma notabilis in Test litter and soil, 1984-1985.

preceded by a few days with lower or more variable temperatures. At the same time, catches of I. notabilis in pit-traps, which are activated on the evening of a routine sampling day, were either greatly increased (Control July 1-2), or decreased (zero captured in Test July 29-30).

The extreme fluctuations we see, then, particularly in leaf litter, may reflect behaviorally induced short-term migrations, overlaid by possibly broad seasonal trends in climatic or substrate preferences. We believe that the point in time at which these samples are taken needs to be characterized in terms of possible causative climatic events. Reliable records of environmental variables are available for 1986 and for much of 1985 (temperature and RH). We propose that barometric pressure, in conjunction with air and soil surface temperature, may also be a parameter necessary for explaining population changes, since Zettel (1984) has demonstrated that activity of some Collembola can be greatly influenced by pressure changes over a time span of only a few hours.

Other species which occur frequently enough in samples for estimating their abundance are few. In soil, only the numerous Onychiuridae qualify, but they cannot be discussed until floatation- and Tullgren-extracted samples are both completely identified (1986 samples). In litter, S. henshawi, O. hexfasciata and T. flavescens are the most common representatives of surface-dwelling families. Tomocerus flavescens and O. hexfasciata, abundant in Test, are not particularly useful for site comparison (Figs. 10, 11), except for their frequency in pit-trap samples in both sites (see section IV. 2. iii.). Sminthurinus henshawi (Fig. 12) exhibited major fluctuations in Control in 1985, possibly in response to unusual temperature changes. All three species, however, reinforce the considerations and arguments brought out above. Especially in 1985, when Collembola populations in general were more abundant than in 1984, fluctuations over time seemed to be more

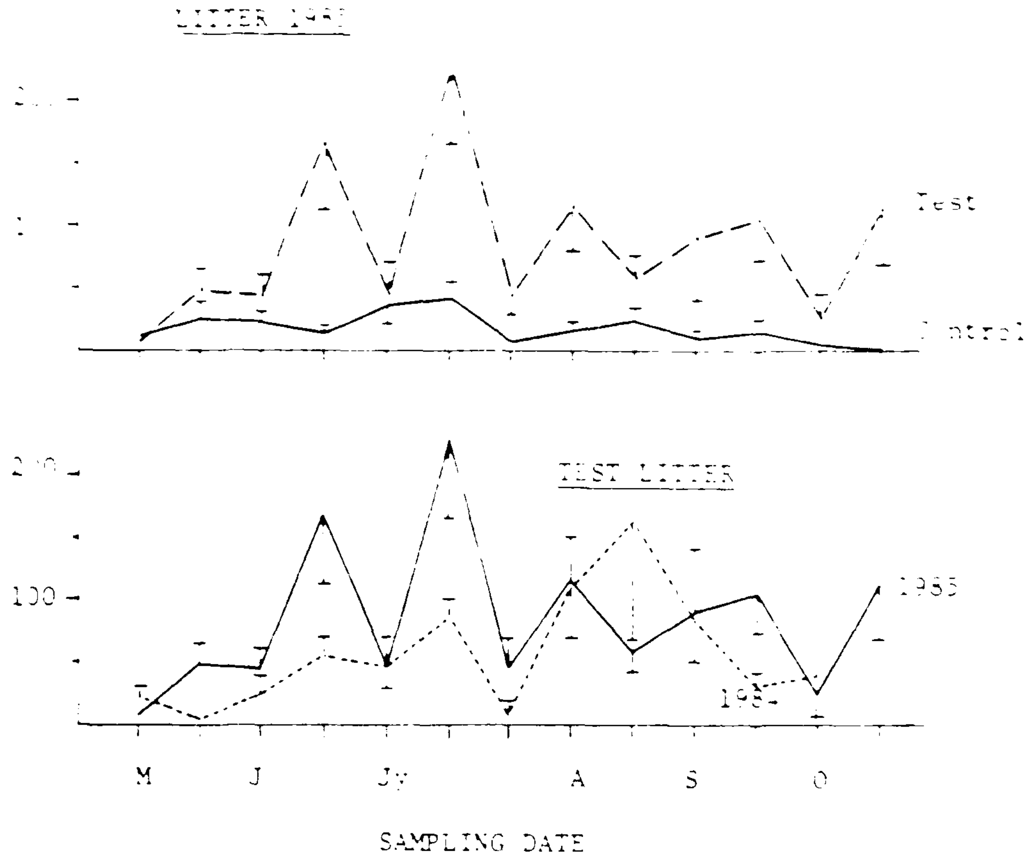

Fig. 1?. Average densities /m^2 $\pm$ SE of Tomocerus flavescens in leaf litter.

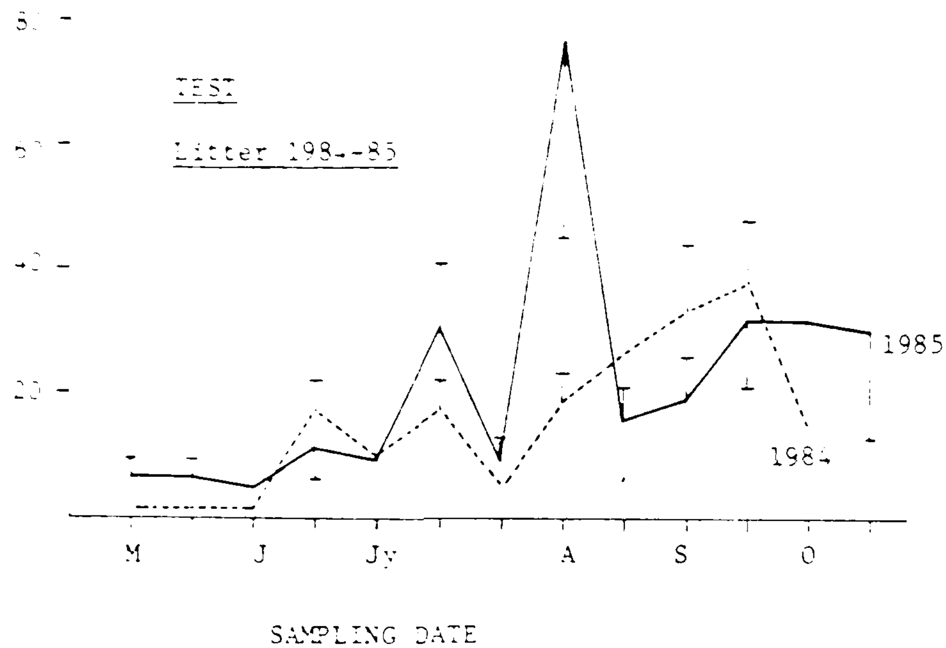

Fig. 11. Mean densities / m^2 ± SE of <u>Orchesella</u> <u>hexfasciata</u> [illegible]

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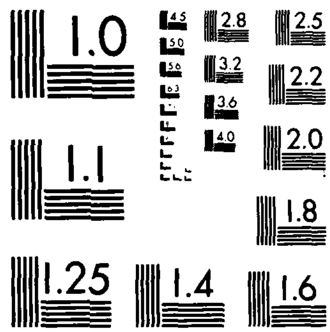

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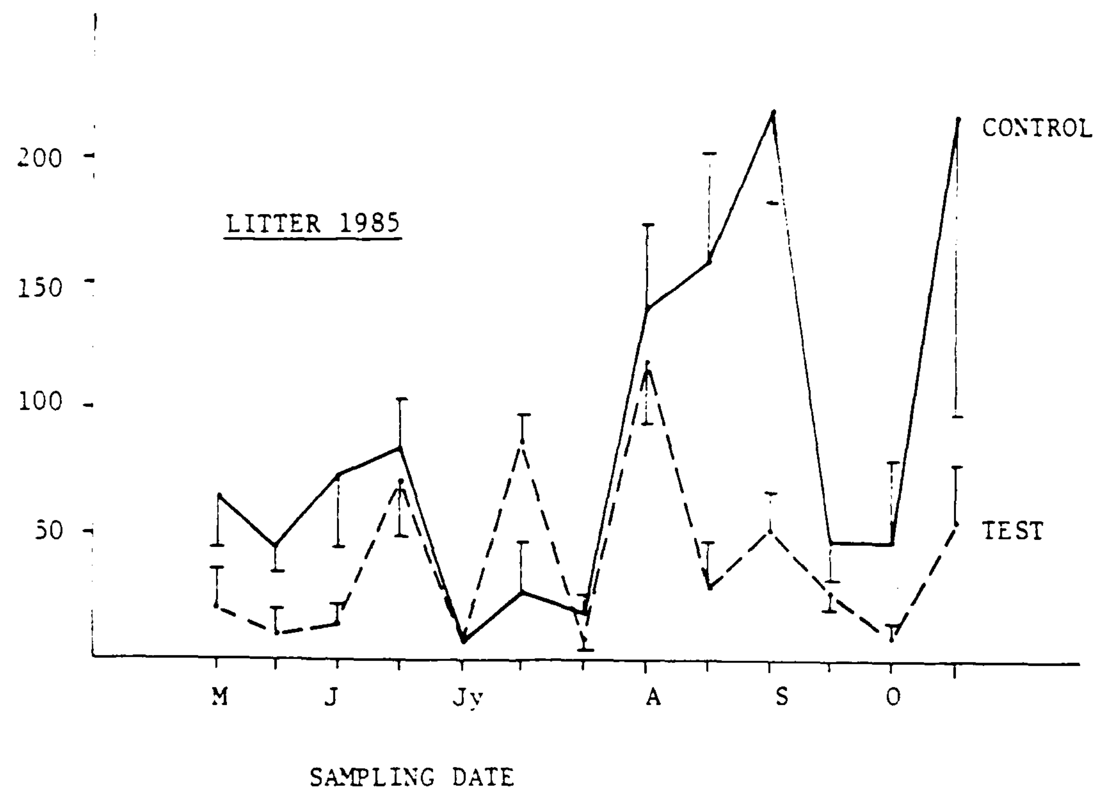


Fig. 12. Average densities/m^2 $\pm$SE of Sminthurinus henshawi in Test and Control, 1985.

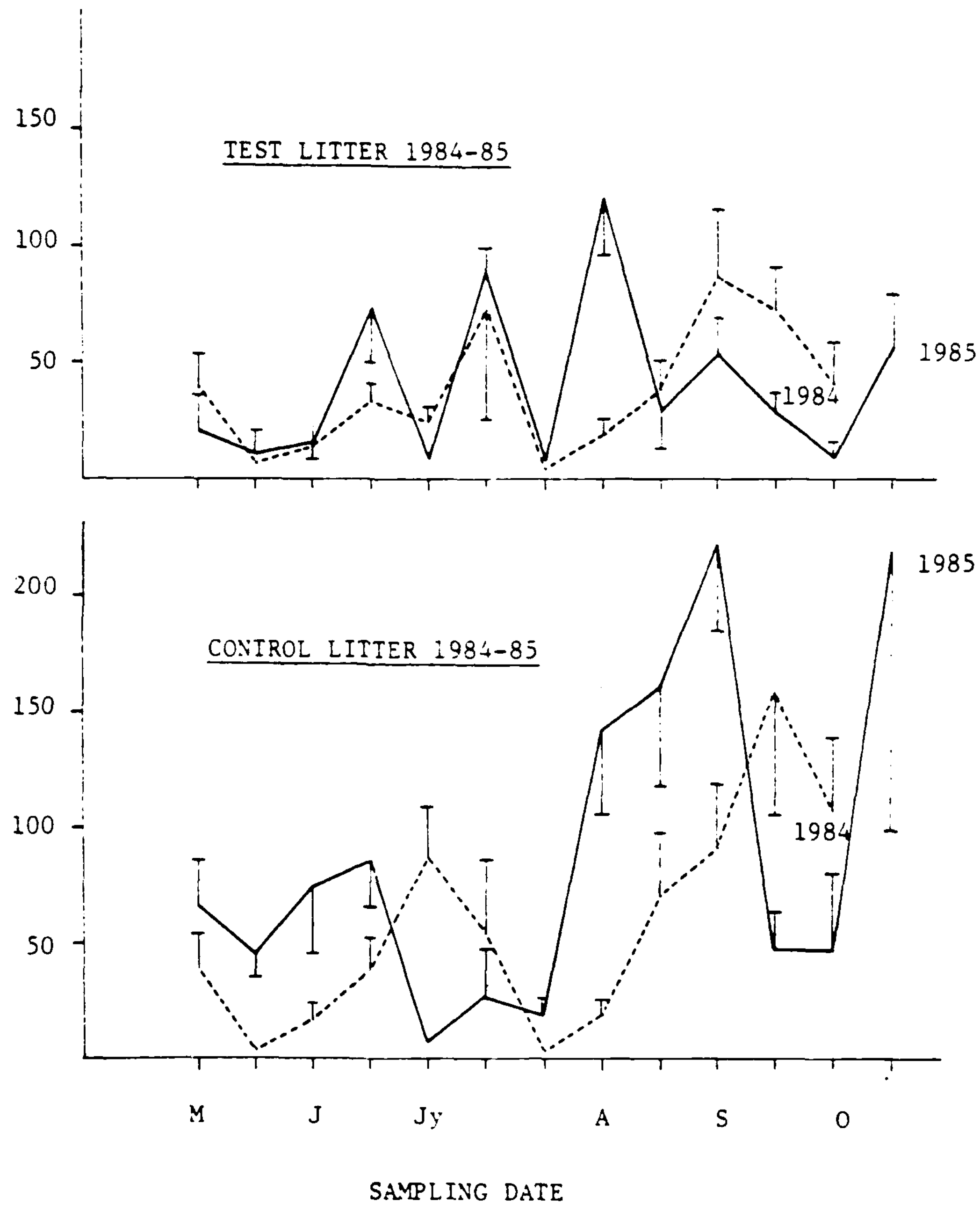


Fig. 13. Mean densities/m^2 ± SE of <u>Sminthurinus</u> <u>henshawi</u> in Test and Control litter, 1984 and 1985.

similar between different species than those within the same species over two consecutive years (Figs. 8, 9, 10, 11, 13, for litter populations).

4. Acari

i. Abundance of major taxa

Average annual density of major taxa decreased from 1984 to 1985 in both sites (significantly so, at $P < 0.05$, only for Mesostigmata in Control) (Fig. 14). As in 1984, major soil-dwelling groups fluctuated relatively synchronously within each site, and population increases in July, August-September and October of 1985 occurred in both sites, although they were not of equal magnitude (Figs. 15-16).

Until we succeed in determining the dominants within Rhagidiidae and Eupodidae, we report their seasonal number at the family level. In both families, abundances followed similar trends in Test and Control (Figs. 17-18), and although densities appeared discrepant, the differences were generally not significant (95% CL overlap). Year-to-year variability within each site showed seasonal differences as great as those between the two sites (Figs. 19-20). Species population analysis will clearly be needed to determine the potential usefulness of these taxa to future conclusions.

ii. Species population dynamics

At the species level, we can now present data on one mesostigmatid and two prostigmatids, selected because they were among the most frequently extracted species in 1983 and 1984.

Three species of Nanorchestes occur in both sites, *N. gilli* and two undescribed species. The most common in both sites is "*Nanorchestes* A", which frequents soil as well as litter, but shows highest frequency in litter samples. As illustrated in Fig. 21, highest population estimates were obtained for fall of 1983. During the following two years, populations were generally more sparse in

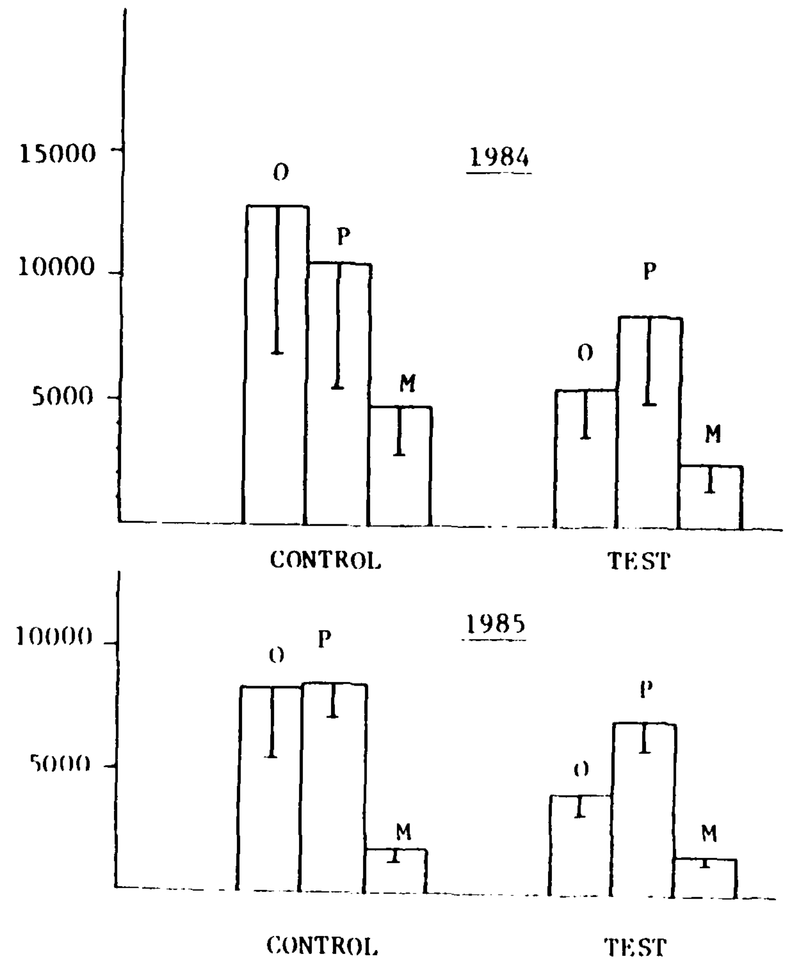


Fig. 14. Average annual densities/m^2 $\pm$ 95% CL, (litter + soil estimates combined), of Oribatida (O), Prostigmata (P) and Mesostigmata (M) in Test and Control, 1984-85.

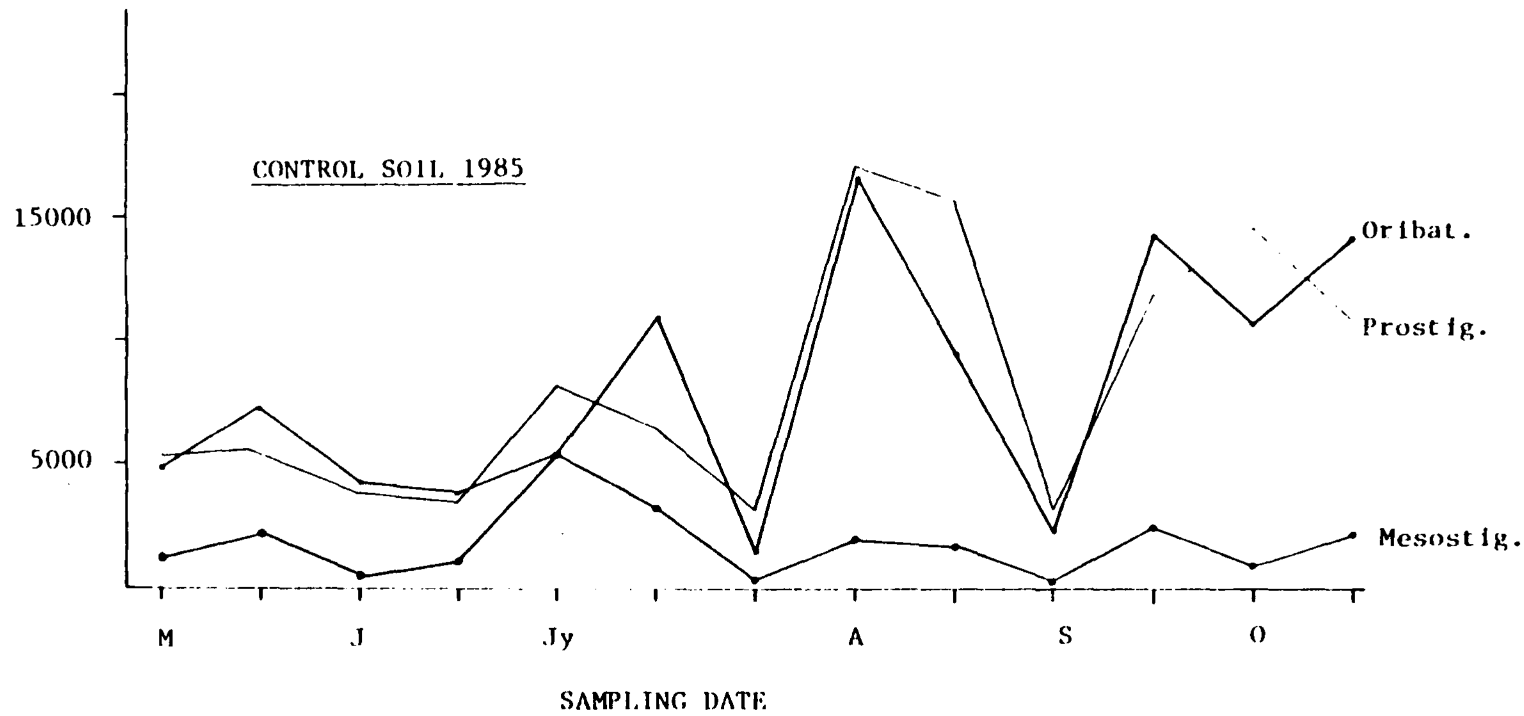


Fig. 15. Average densities of major mite taxa in Control soil, 1985.

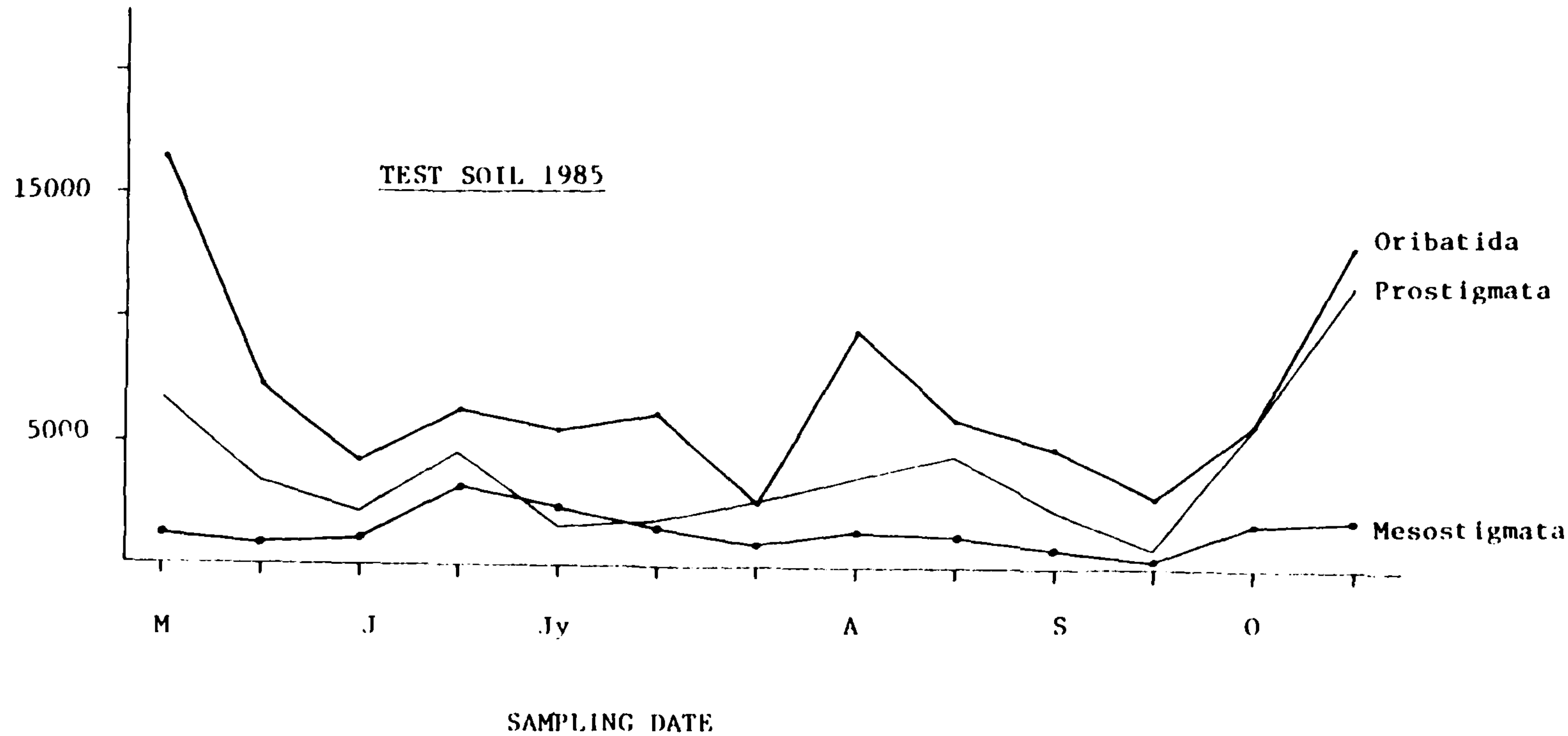


Fig. 16. Densities/m^2 of major mite taxa in Test soil, 1985

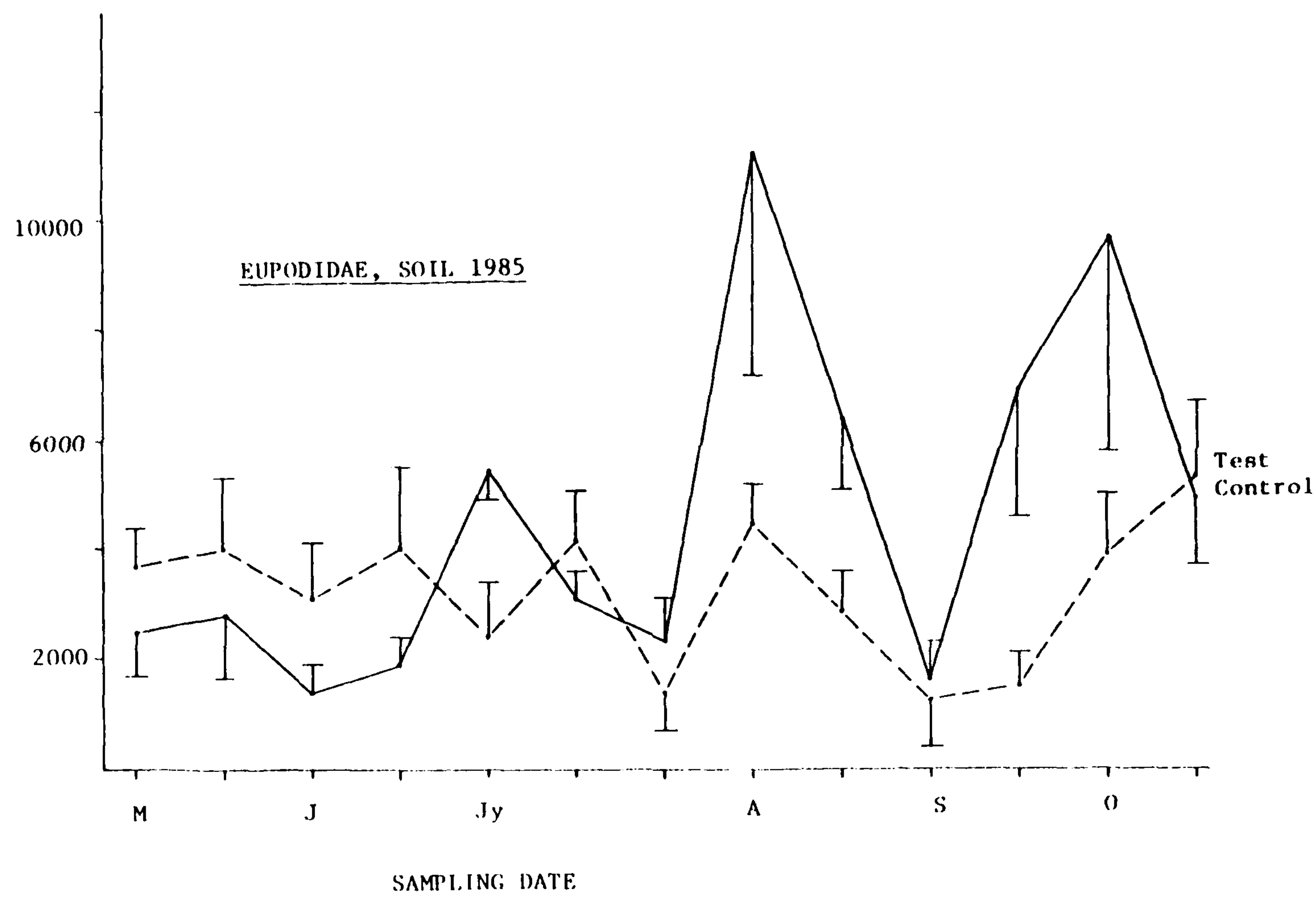


Fig. 17. Densities/m^2 $\pm$ SE of Eupodidae in Test and Control soil, 1985.

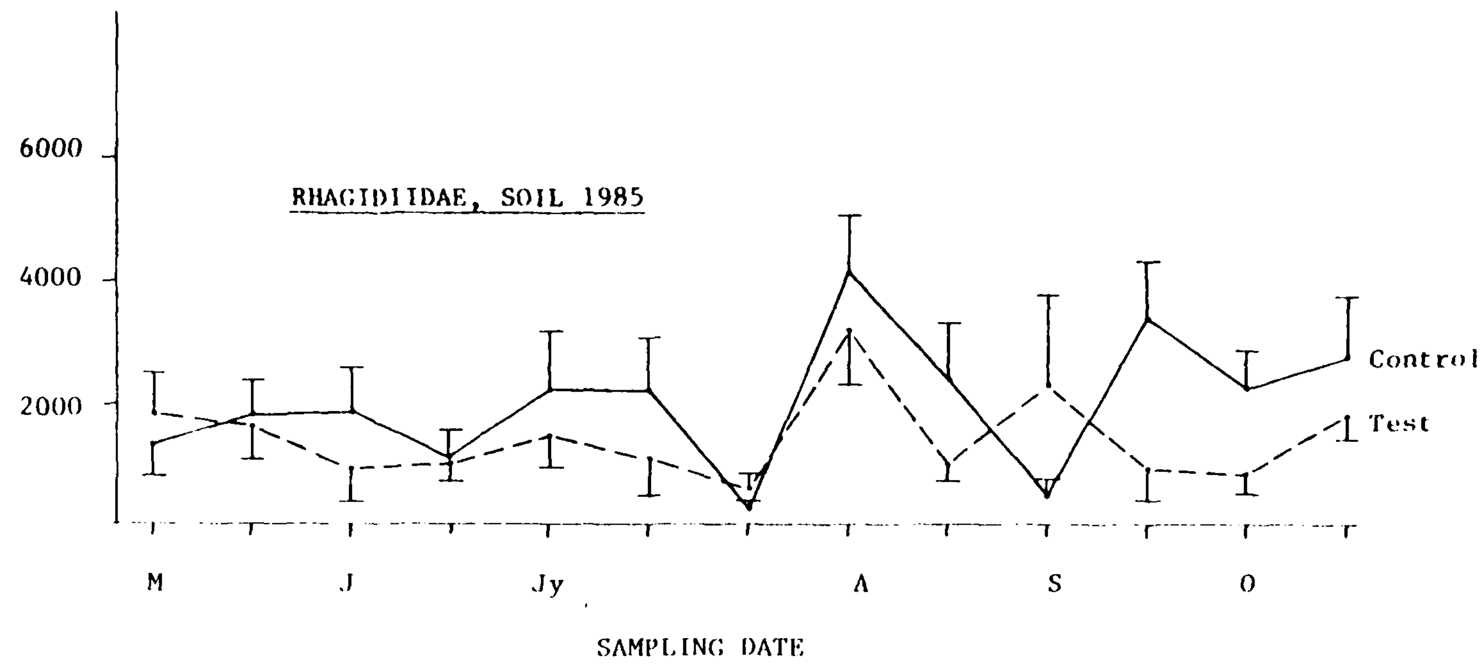


Fig. 18. Densities/$m^2 \pm$ SE of Rhagidiidae in Test and Control soil, 1985.

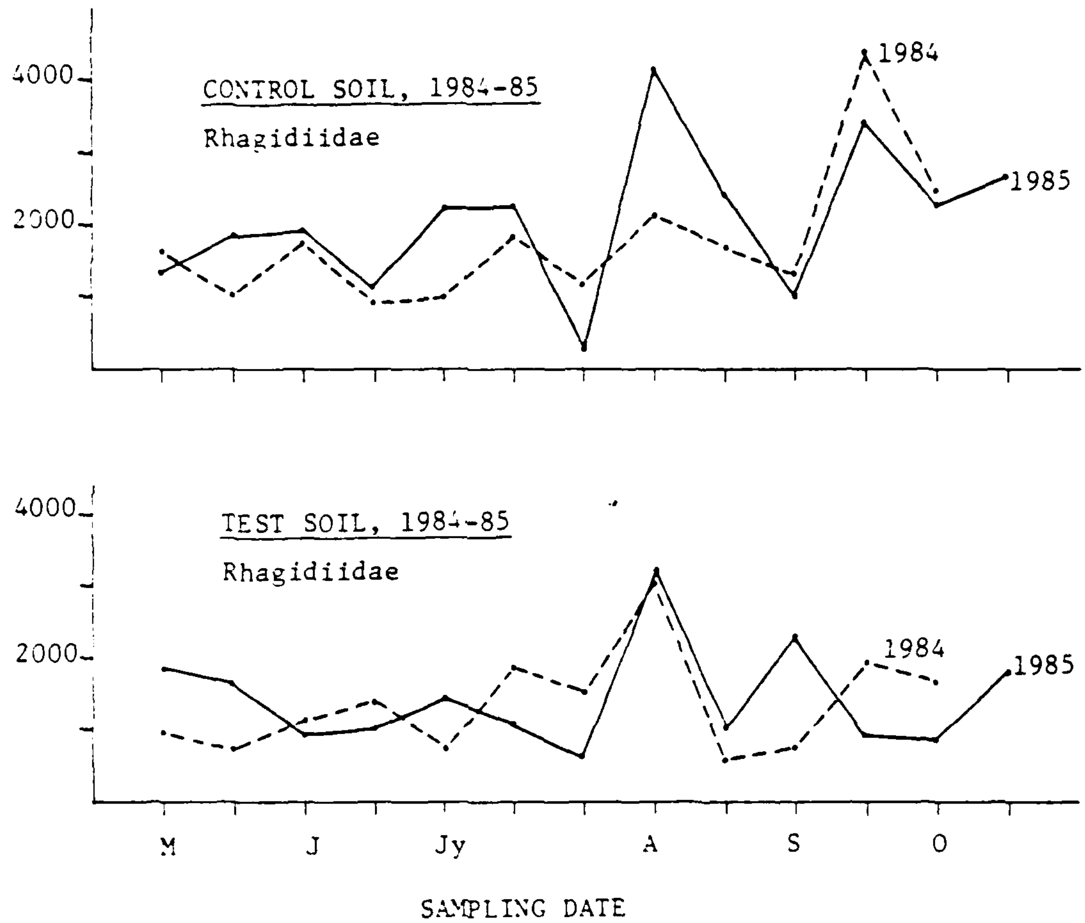


Fig. 19. Densities/ m^2 of Rhagidiidae in soil, 1984 and 1985.

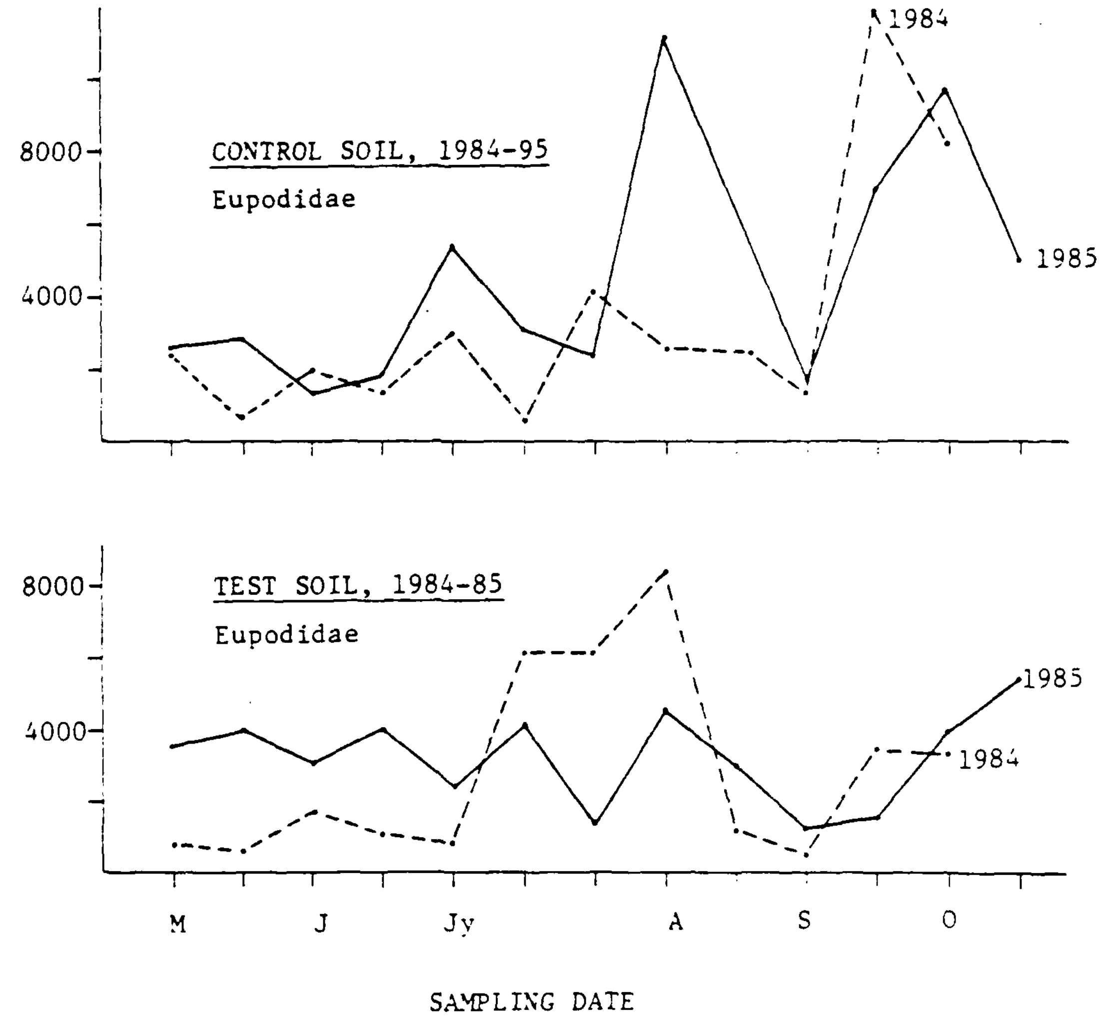


Fig. 20. Densities / m^2 of Eupodidae in Soil, 1984 and 1985.

both sites, with maxima and minima alternating somewhat randomly. In both sites, gravid females were present during every month from May through September, with the exception of August and September in Test 1985: the very low numbers extracted from litter at that time (Fig. 21) probably masked their presence by sheer chance. Year-round reproduction, with peaks in May through August, appeared to be the rule in the species.

The year-to-year decline in numbers of Nanorchestes A was observed in Asca aphidioides (L.) as well. In both sites (Fig. 22), 1985 densities were well below 1984 estimates and declined essentially to zero in early September. Populations peaked in June-July and August as a consequence of the appearance of larvae and the increasing abundance of nymphal stages thereafter. Apparent population declines may not have been a function of abundance, but rather of an increasing proportion of molting or newly molted immatures in the population (resulting in lower extraction efficiency). The life cycle of A. aphidioides was clearly univoltine in both sites, as illustrated in Figs. 23 and 24: the species overwinters in the adult stage (males are unknown), and consists mainly of gravid females in May and June. Larvae hatch in June through August, and undergo development through proto- and deutonymphs during summer and early fall, closing the cycle with a late-autumn population consisting entirely of adult females. This developmental cycle seemed prolonged in 1984, more rapid and tight in 1985. Whether cumulative temperatures differed enough between years to have had an effect on developmental rates needs to be analyzed.

The mesostigmatid "Species A" exhibited a year-to-year decline similar to those discussed above, and discrepancies between Test and Control were relatively pronounced (especially in 1984, Fig. 25). The yearly cycle of reproduction and recruitment (Figs. 26, 27) was similar to that of A. aphidioides, but less well defined: a small proportion of gravid females occurred in late fall, and larvae

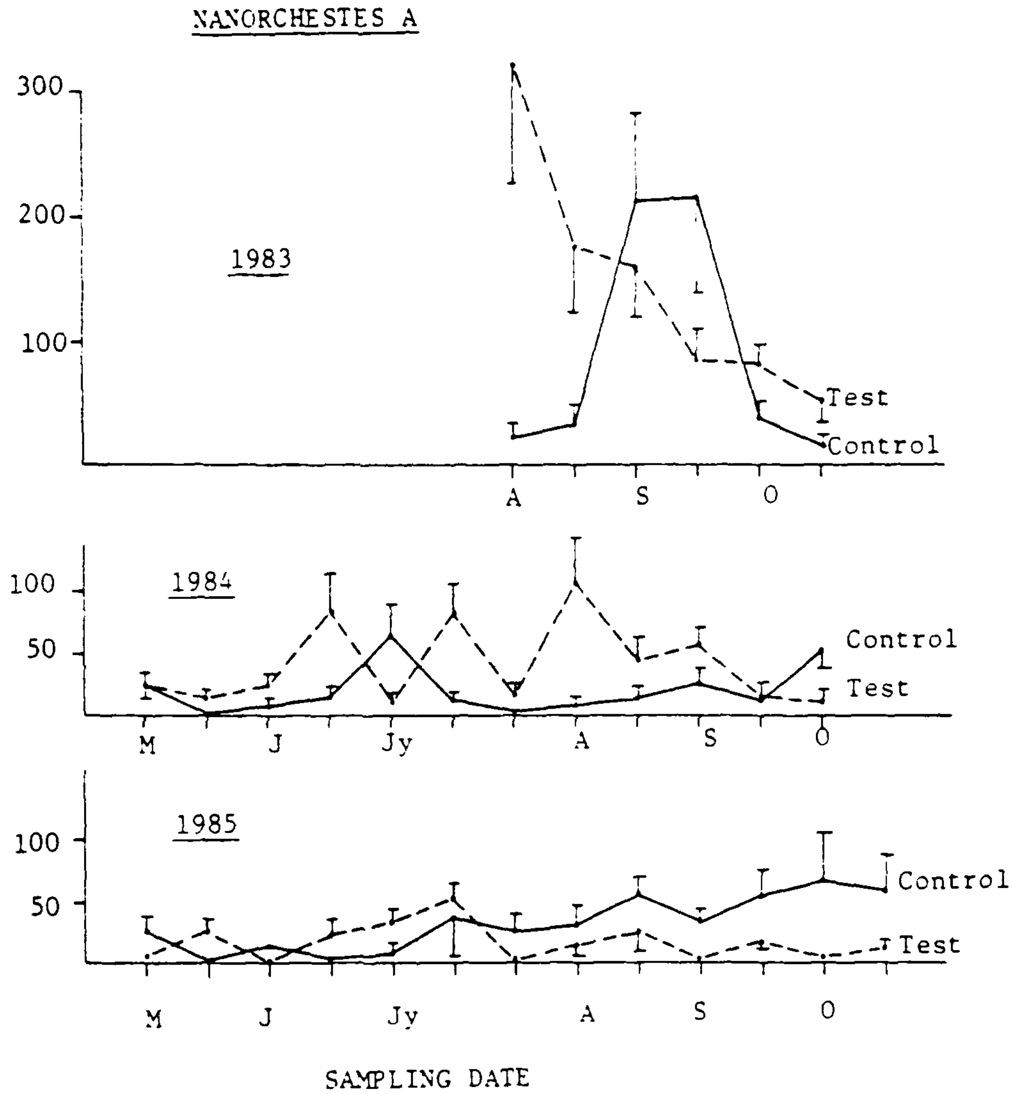


Fig. 21. Densities/m^2 ± SE of Nanorchestes sp. A in Test and Control, August 1983 to October 1985.

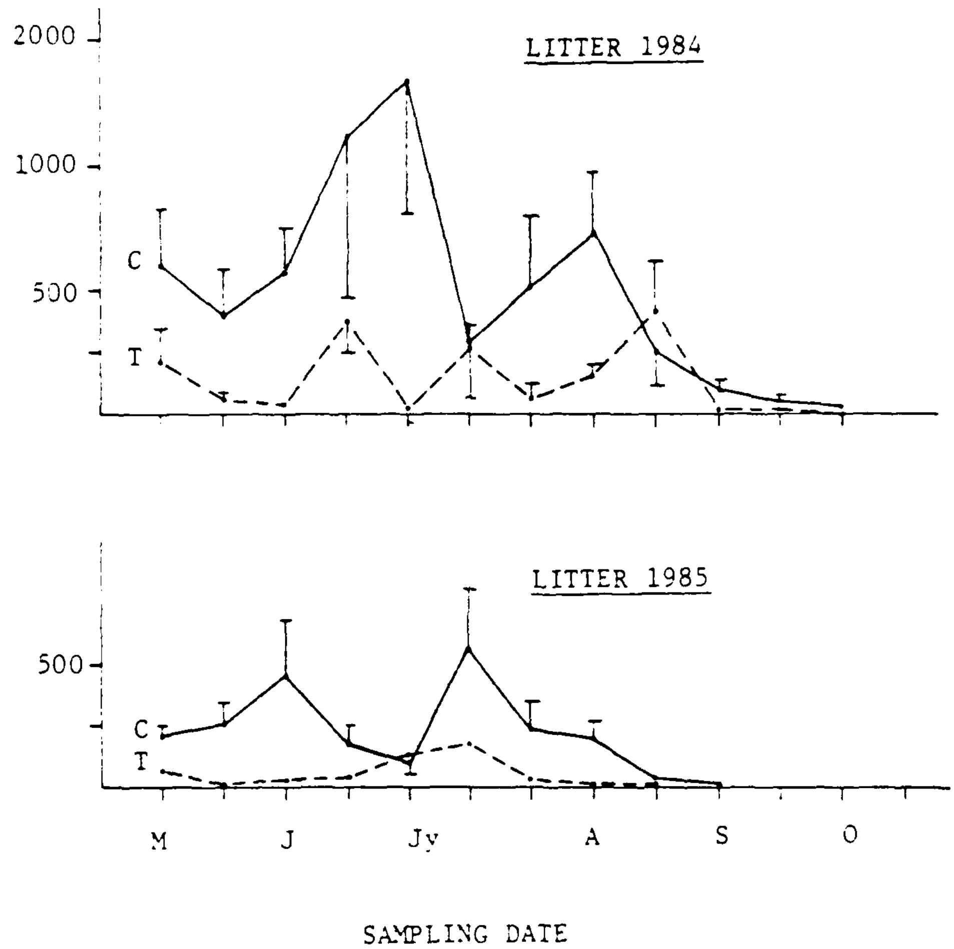


Fig. 22. Densities/m^2 $\pm$ SE of <u>Asca aphidioides</u> in litter, in Test and Control (T and C) in 1984 and 1985.

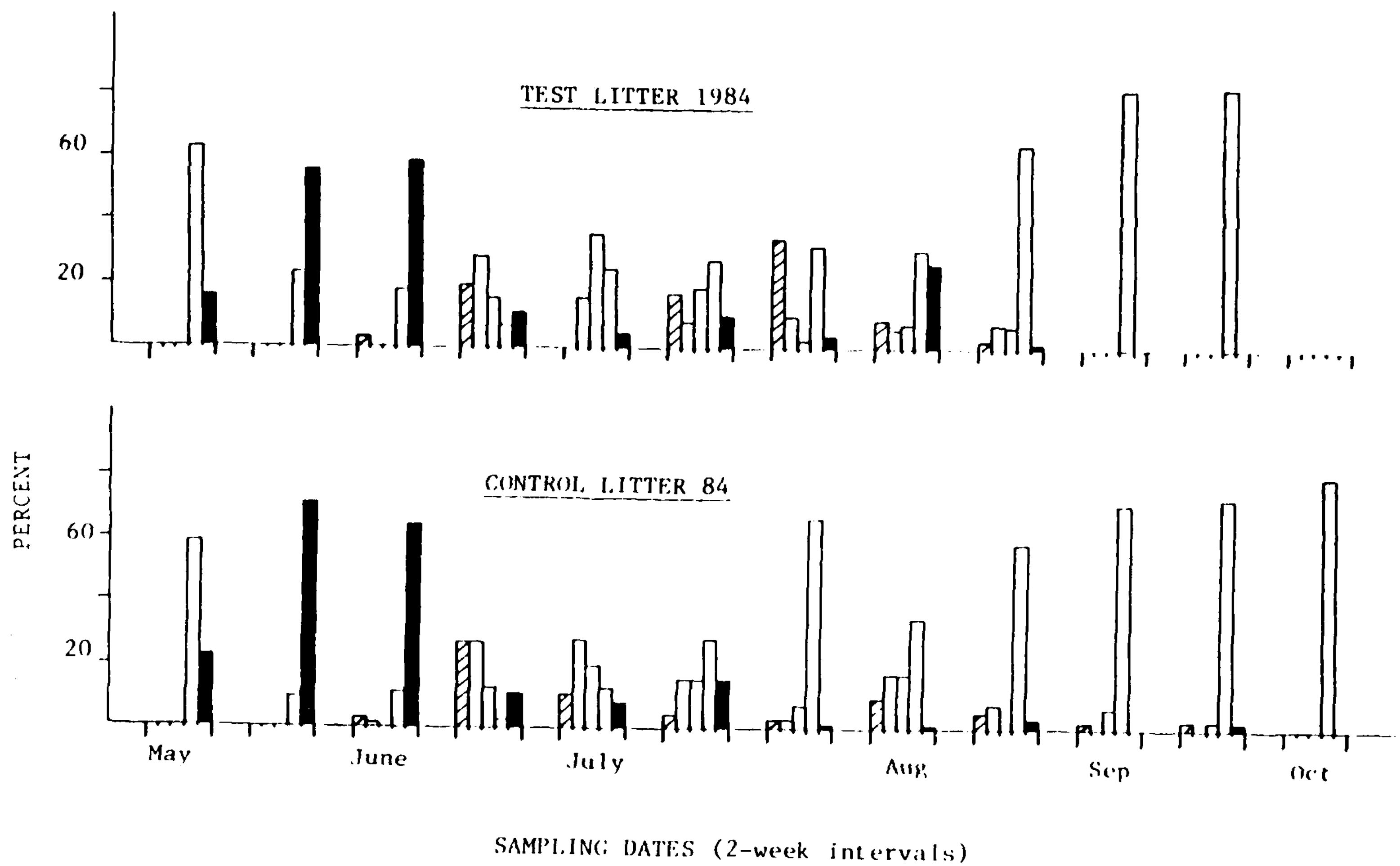


Fig. 23. Asca aphidioides: population stage structure in Test and Control, in % of total N per date. Hatched bars: larvae; black bars: gravid females; open bars from left to right: protonymphs, deutonymphs, and non-gravid females.

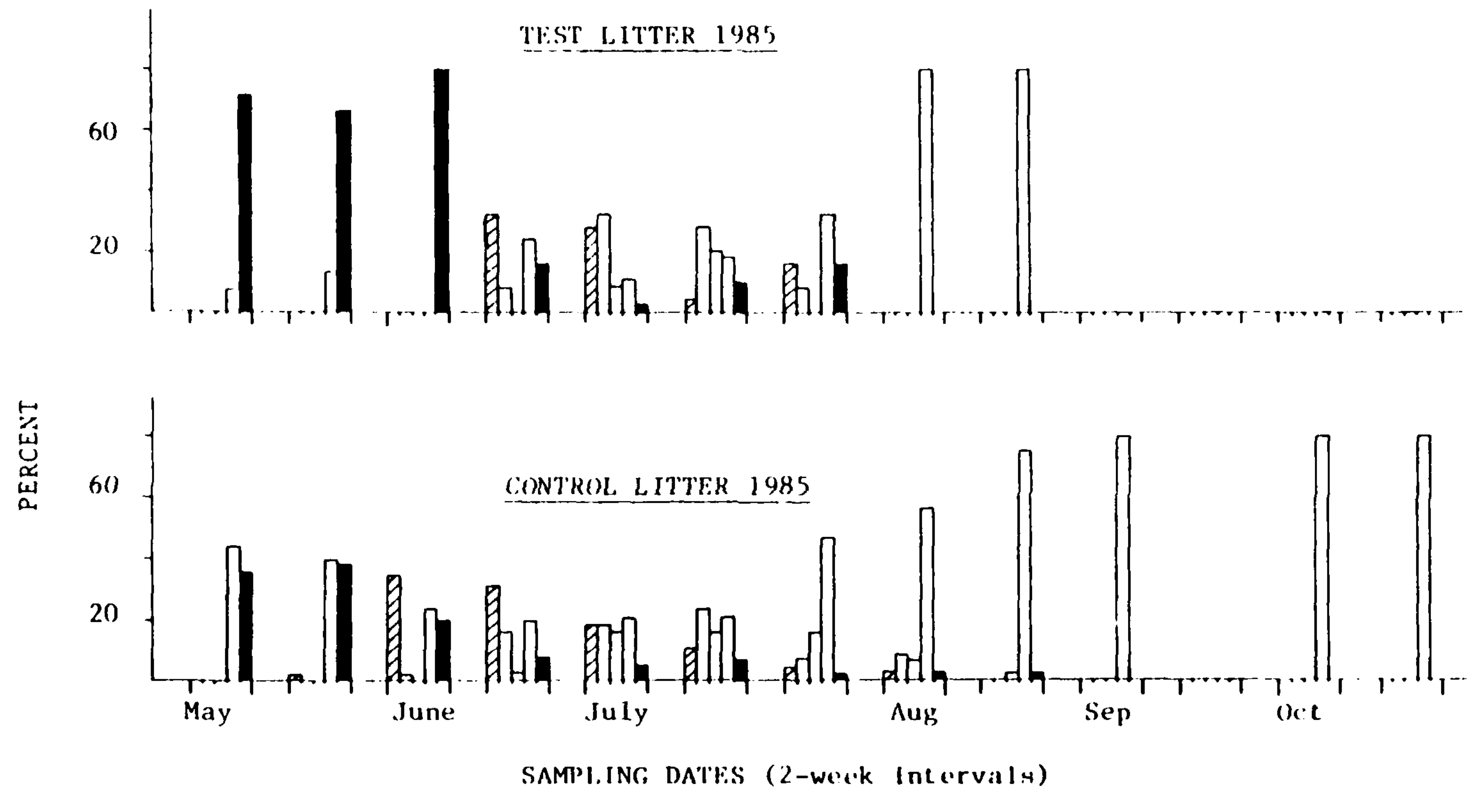


Fig. 24. Asca aphidioides: population stage structure in 1985, in % of total N per date. Hatched bars: larvae; black bars: gravid females; open bars from left to right: protonymphs, deutonymphs, and non-gravid females.

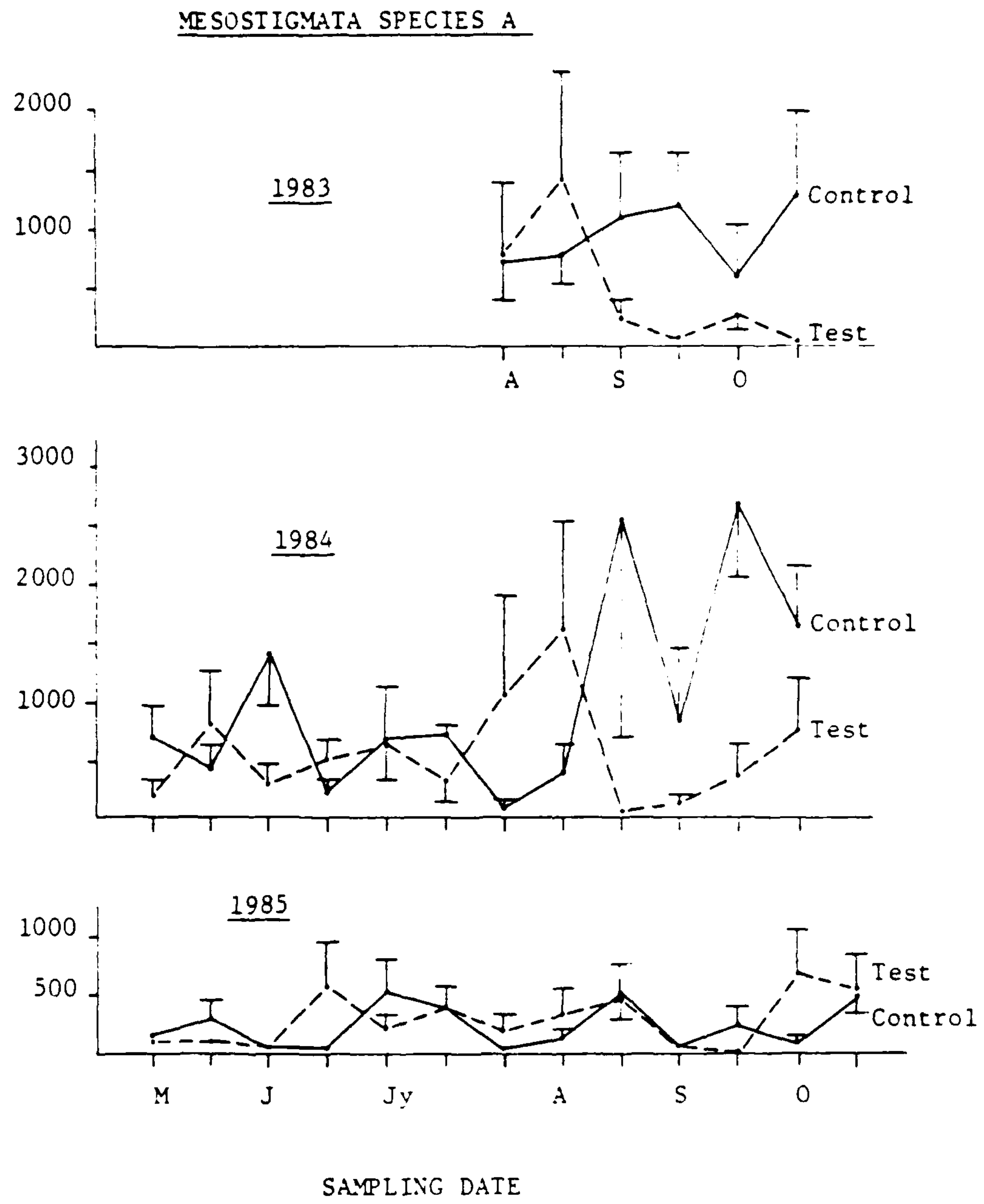


Fig. 25. Densities/m^2 $\pm$ SE of the mesostigmatid "Species A" in Test and Control soils, August 1983 to October 1985.

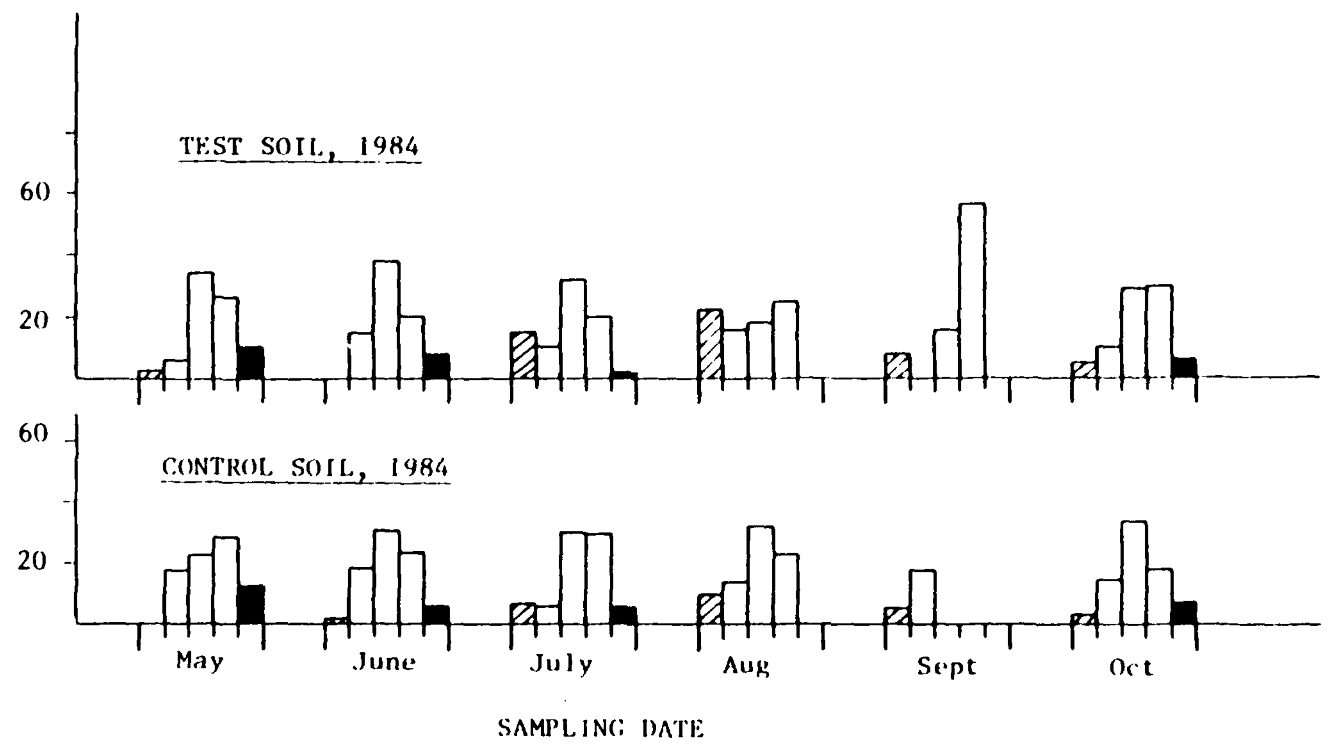


Fig. 26. Population stage structure of Species A (Mesostigmata), in % of total N, summed by month, 1984. Hatched bars: larvae; Black bars: gravid females; open bars from left to right: protonymphs, deutonymphs, non-gravid females.

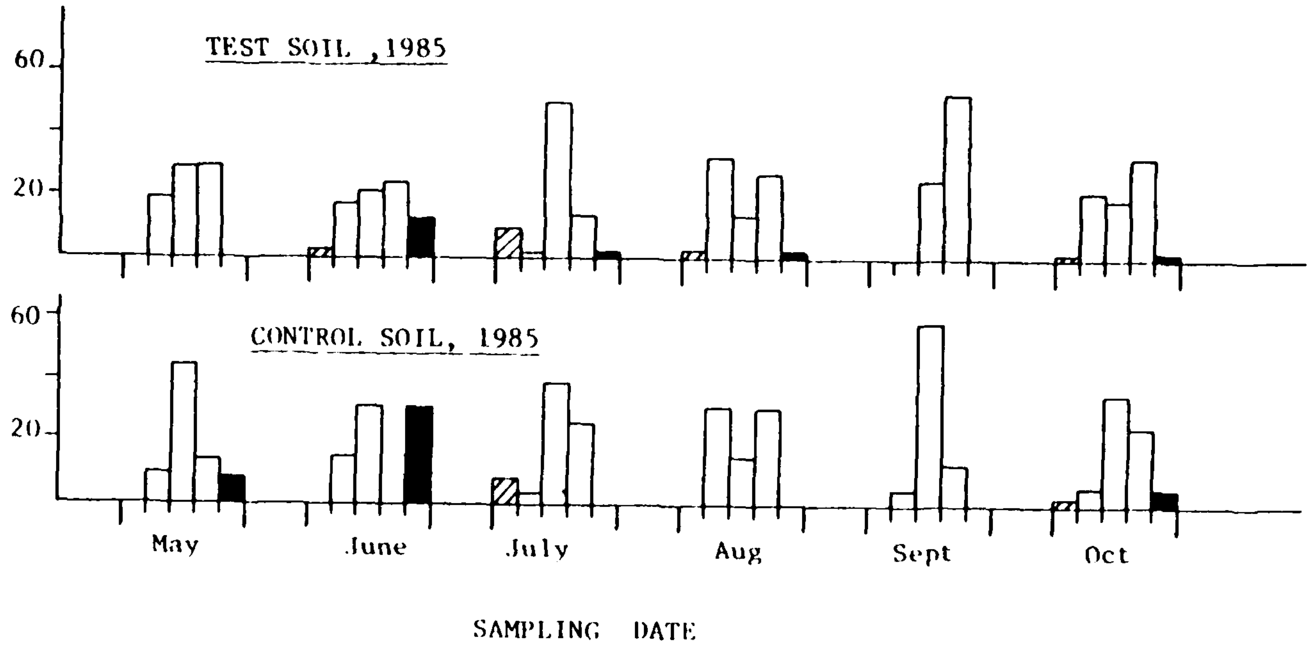


Fig. 27. Population stage structure of Species A (Mesostigmata) in Test and Control soil, expressed in % of total N/ month. Hatched bars: larvae; black bars: gravid females; open bars from left to right: protonymphs, deutonymphs and non-gravid females.

were occasionally present in the spring (Fig. 26). All developmental stages overwintered, although July and August were the peak months of recruitment in both sites.

5. Statistical treatment

In the case of litter-dwelling species, density data indicate that some correlation between numbers of animals and environmental factors may exist. As a first step, the significance of regression of counts on temperature and moisture, chosing a relatively high oc (say, 0.3) will determine which variables are important enough to use as covariates. Densities can then be tested in a General Least Squares program (ANOVA with appropriate covariates).

Since sampling dates correspond to each other from year to year, we also anticipate using means/date lumped over years; to give us three years of data, we need to wait until 1986 density data become available.

For soil-dwelling species, lumped means of N/m^2 over years provide an appropriate data base for analysis of variance to test site and date effects and their interactions.

All density data need to be log-transformed for use in parametric statistics.

Seasonal stage structures (frequencies): data are first lumped into overall yearly totals, to test whether numbers in Test and Control are proportionally the same (contingency tables, for instance, 2 sites x 3 years). Expanded contingency tables are then used to test detailed distributions of numbers/stage/date.

IV. SURFACE-ACTIVE ARTHROPODA

1. Methods and status of data

Since the beginning of 1985, pit-traps have been provided with four (1 m) barriers shallowly embedded at right angles to each other and abutting to the rim of standard pit-traps. For certain taxa, e.g. carabids and spiders, barrier-trapping resulted in greatly increased catch sizes. At this time, data on Carabidae, Collembola and velvet mites are available. Determination of the ample spider material nears completion, but we defer the pertinent account until the second draft of this report.

Sorting of 1986 catches is nearing completion, and identification of species has begun.

2. Collembola

i. Barrier effects

As reported earlier, a preliminary experiment during 1984 indicated that barriers could be expected to yield approximately doubled numbers of captured Collembola. Since captures are likely to be density-dependent, a barrier effect might well be confounded with the influence of population density. Of particular relevance here is the overall increase in colle   populations from 1984 to 1985 (Section III.2.).

Expressed as a simple multiplication factor, changes in density and changes in trap catches are summarized in Table 10: e.g., while total Collembola populations in both sites increased by a factor of 1.4, trap catches were higher by 1.8 in Control and 2.3 in Test. Among the dominant families, Sminthuridae were virtually not affected at all by barriers, catches being almost unchanged from 1984 to 1985, and density increases higher than capture increases. The reverse was true of Entomobryidae and Isotomidae, in which capture were augmented by barriers well beyond abundance increases in the same year. Each species (only dominants

are shown in Table 10) was affected differently, and no consistent proportionality between densities and catch sizes existed. Analysis of possible relations, and their year-to-year constancy, between trap catches and abundance will be meaningful only once 1986 (second barrier-trap year) data are at hand.

Table 10. Changes in mean annual population density (N/m^2, litter + soil), and in trap catches of dominant families and species, from 1984 to 1985; x factor = N 1985 / N 1984.

	x FACTOR FOR:			
	TEST		CONTROL	
	N/m^2	Traps	N/m^2	Traps
Sminthuridae	1.8	1.2	2.0	1.0
S. henshawi	1.7	1.0	1.6	0.9
Entomobryidae	1.3	4.2	0.9	3.0
T. flavescens	2.0	5.9	1.5	1.9
O. hexfasciata	1.8	3.1	0.8	2.8
Isotomidae	1.5	2.1	1.4	2.3
I. notabilis	1.4	5.3	1.8	4.7
Total Collembola	1.4	2.3	1.4	1.8

ii. Species composition and diversity

Trapping in 1985 yielded essentially the same species as in 1984, with addition of three in Control and five in Test which had not yet been recorded and were captured in low numbers. Keeping in mind that year-to-year density fluctuations as well as barriers exerted their effects on apparent community structure, Table 11 summarizes dominance values at the family level. Clearly, they were altered considerably. Sminthurid prevalence in 1984 was reduced in 1985 by the

relative ineffectiveness of barriers (Table 10) on one member of this family. Entomobryid dominance increased markedly in Test, mainly due to barrier-augmented (about 6-fold, Table 10) catches of the already abundant dominant I. flavescens. An unexpected rise of hypogastrurids in Control (Table 11) was traceable to a single species, P. saxatilis, which exhibited unprecedented activity peaks in July and August of 1985.

The same considerations apply to the most frequently captured species, for which Table 12 lists dominance values in both years.

Table 11. Percent dominance of surface-active collembolan families in 1984-85.

	% OF TOTAL N			
	TEST		CONTROL	
	1984	1985	1984	1985
Sminthuridae	46.1	21.0	63.2	36.1
Entomobryidae	45.4	73.2	20.7	35.0
Isotomidae	6.4	5.1	5.5	7.6
Hypogastruridae	2.1	0.7	10.6	21.3
Total N	4,443	11,518	5,068	9,946
N species	31	36	27	30

Table 12. Percent dominance of common surface-active Collembola, Test and Control 1984-85.

	TEST		CONTROL	
	1984	1985	1984	1985
S. henshawi	35.8	14.2	53.1	26.2
S. lepus	7.4	5.8	4.2	4.0
T. flavescens	16.0	36.6	8.1	8.5
O. hexfasciata	23.4	27.8	7.2	11.0
E. nivalis	1.6	4.6	-	-
I. notabilis	-	1.5	1.8	6.2
I. nigrifrons	3.4	1.3	3.0	-
I. viridis	2.3	2.2	-	-
P. saxatilis	1.1	-	10.2	19.4

Again with the confounded abundance-barrier effects in mind, we would expect that diversity indices might change in 1985 (Table 13). For instance, relatively lower catches of *S. henshawi* and larger numbers of *P. saxatilis* and others, resulted in a more than doubled equitability index for Control. H' diversity increased slightly in both sites, as did community similarity taking into account abundance (C_N, Table 13).

Table 13. Community indices for surface-active Collembola in 1984-85, based on total N/ species.

	1984		1985	
	Test	Control	Test	Control
H' diversity	0.84	0.76	0.90	0.93
Simpson dominance λ	0.22	0.32	0.24	0.15
Equitability $(1/\lambda)$	4.55	3.16	4.20	6.78
Bray-Curtis simil. C_N	0.29		0.42	
Sørensen's simil. C_S	0.79		0.79	

iii. Seasonal and diurnal activity

Seasonal catches of the most abundant families are illustrated in Figs. 28 and 29. Clearly, sminthurids and entomobryids as a whole fluctuated synchronously in Test and Control, which is not surprising given the close agreement in air temperature and RH regimes, and the relative concordance of species composition.

Activity patterns of three dominant species common to both sites are illustrated in Figs. 30-32. Sminthurinus henshawi (Fig. 30), as discussed in earlier reports, is a flexible species, switching between diurnal and nocturnal activity in apparent response to temperature. Orchesella hexfasciata, mainly diurnal, exhibited brief periods of nocturnal activity simultaneously in both sites (Fig. 31). Tomocerus flavescens, on the other hand, is strongly nocturnal, day-activity being prevalent on only two dates in 1985 (Fig. 32), both times following a cold night.

All three species exhibited clear overall seasonal patterns: entomobryids were most active during mid-summer, while S. henshawi showed early summer and early fall peaks. All three also yielded data in good between-site agreement, and large enough numbers for analysis; although there are gaps in our environmental data for 1985, statistical treatment will be possible (see section IV.4.).

3. Carabidae

i. Species composition and diversity

The number of species captured increased in 1985 from 16 to 21 in Test and from 18 to 21 in Control, all new records being rare species. Dominance relationships did not change drastically in Test, P. melanarius still leading the community, followed by P. pensylvanicus and P. mutus (Table 14). In Control, greatly increased catches of S. impunctatus brought this species to first rank at 30% dominance, and reduced the relative importance of P. coracinus, P. pensylvanicus and P. adstrictus considerably.

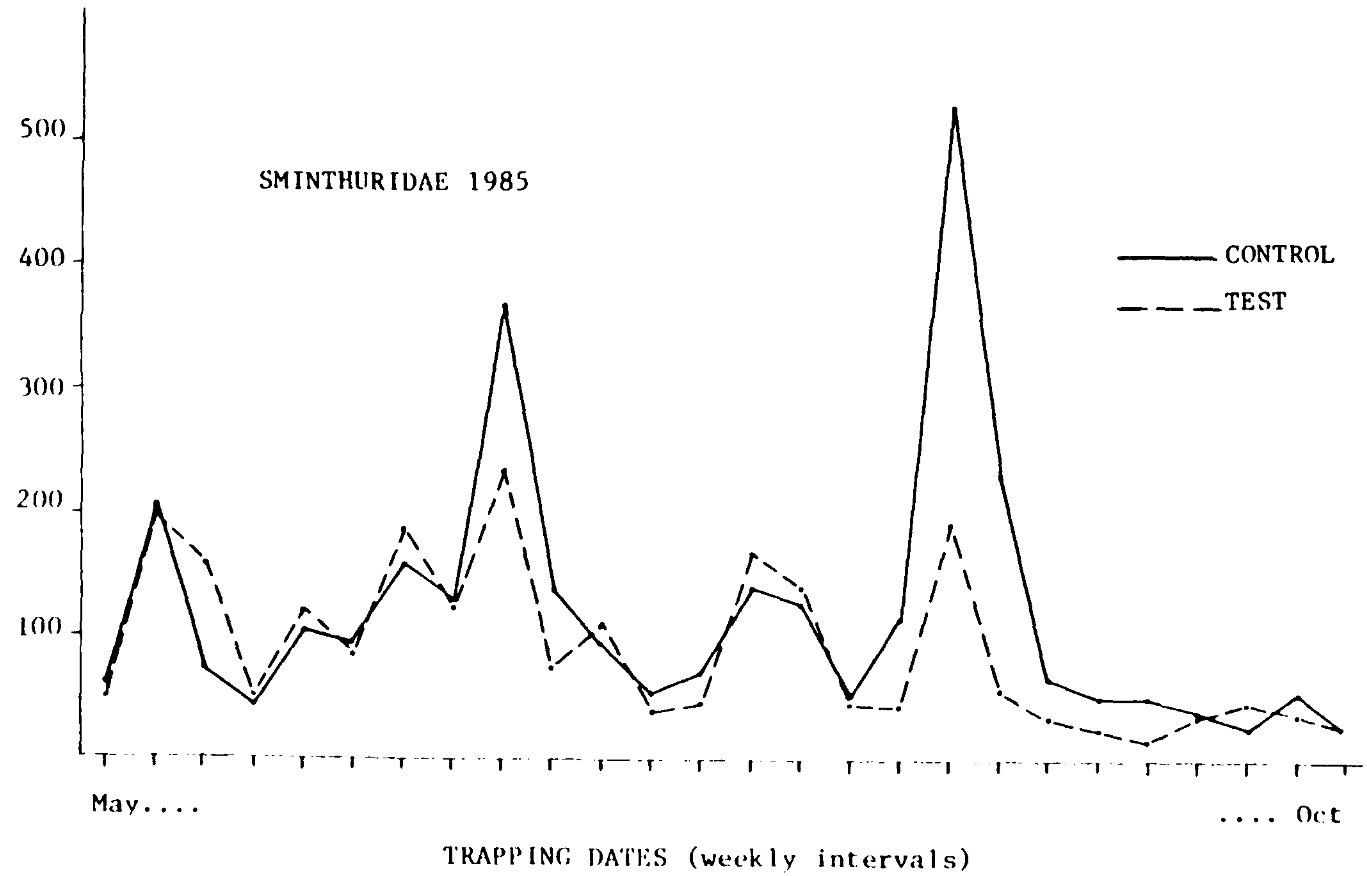


Fig. 28. Total catches of Sminthuridae (night + day) in Test and Control pit-traps, 1985.

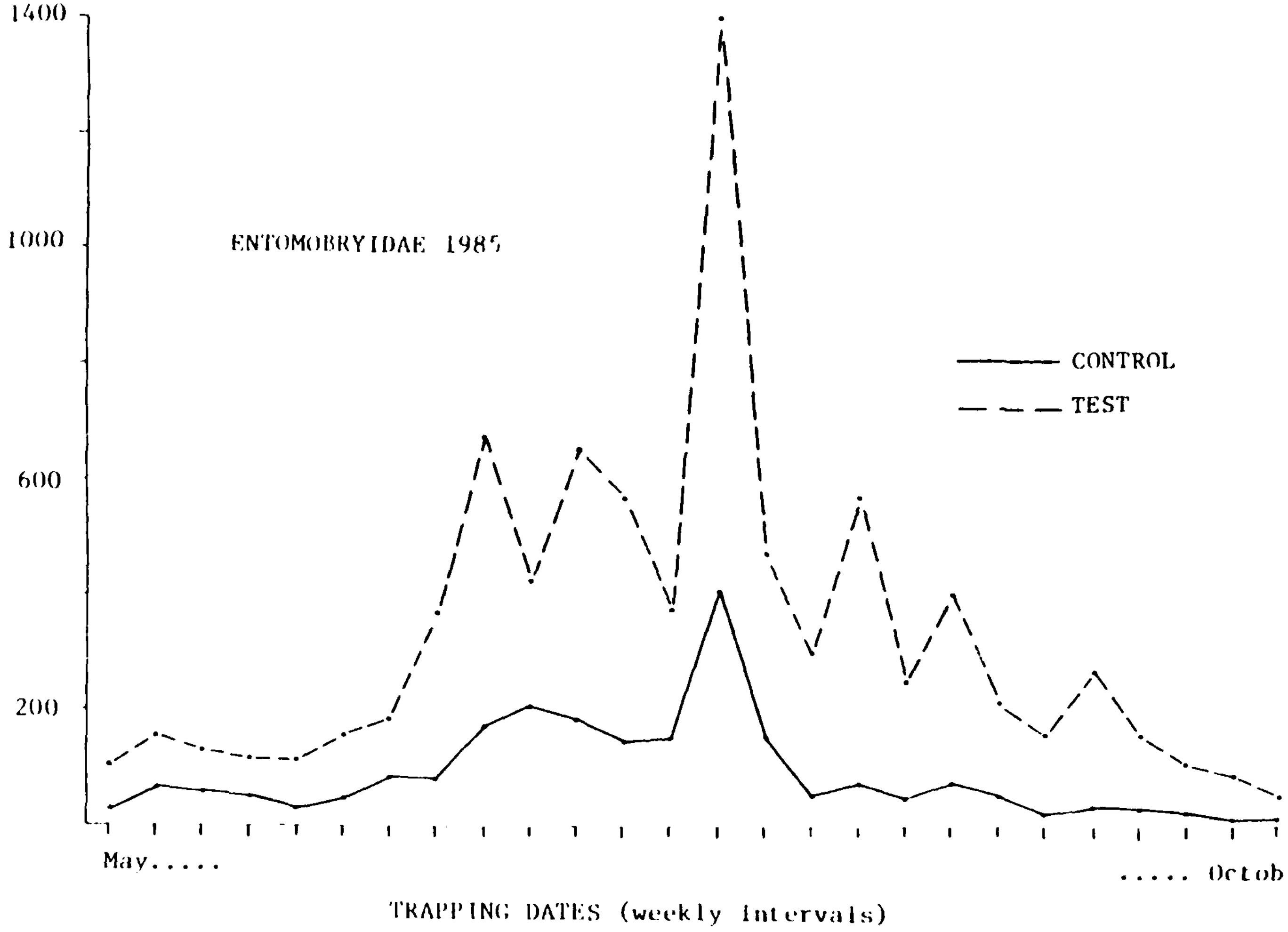


Fig. 29. Total weekly catches of Entomobryidae (night + day) in Test and Control, 1985.

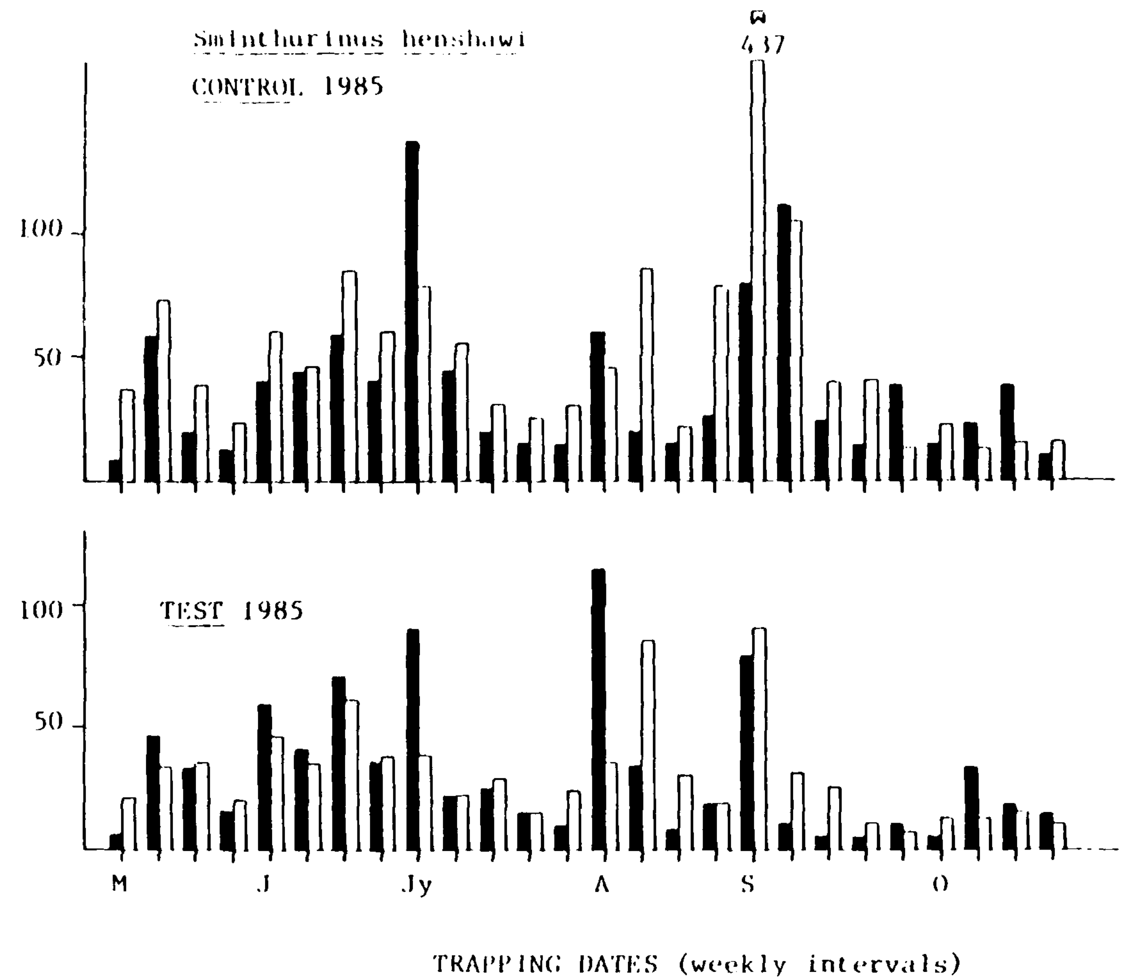


Fig. 30. Total night and day catches of Sminthurinus henshawi in 1985.

Open bars = day, black bars = night.

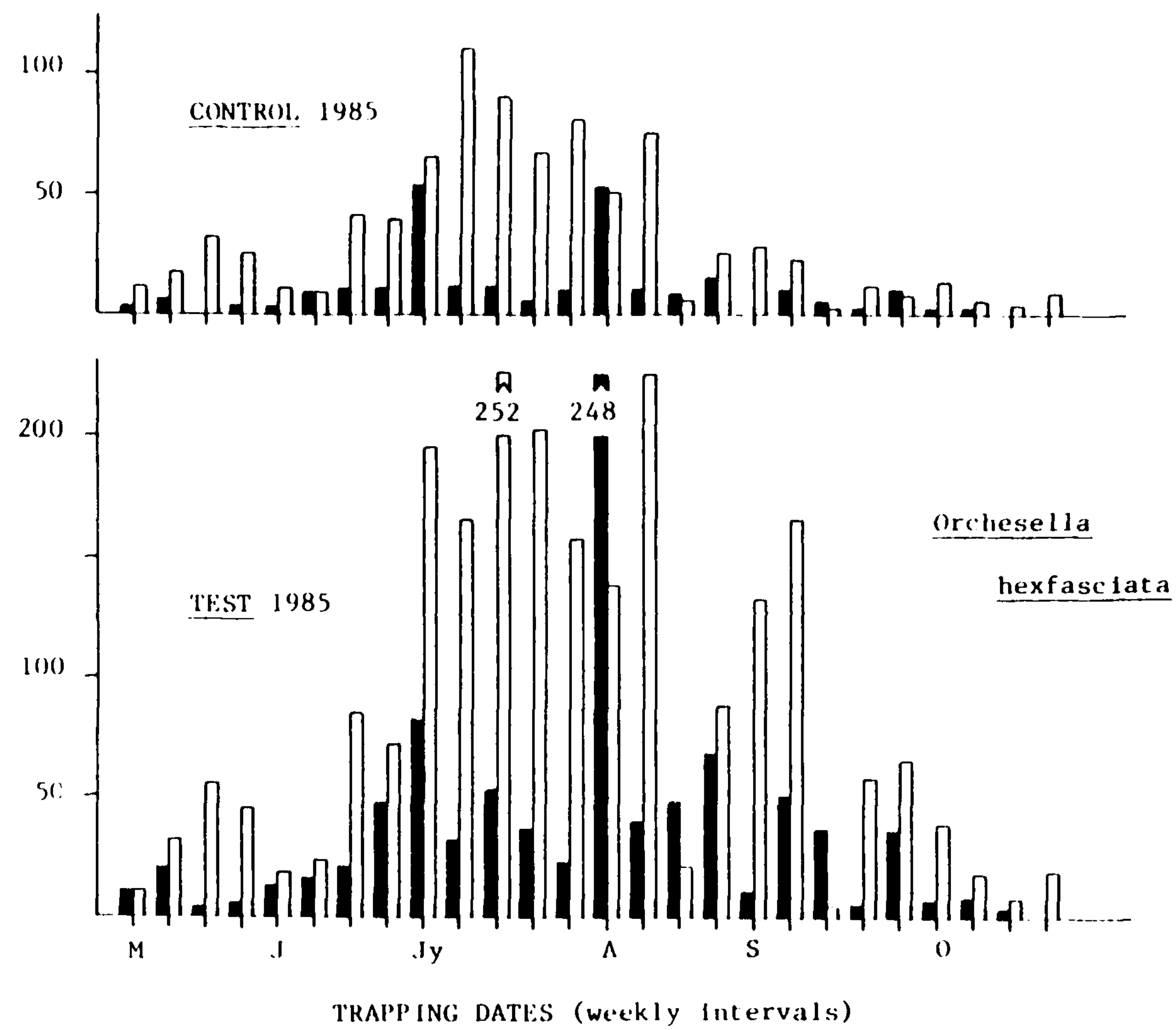


Fig. 31. Total night and day catches of <u>Orchesella hexfasciata</u> in 1985 in Test and Control. (open bars = day, black bars = night).

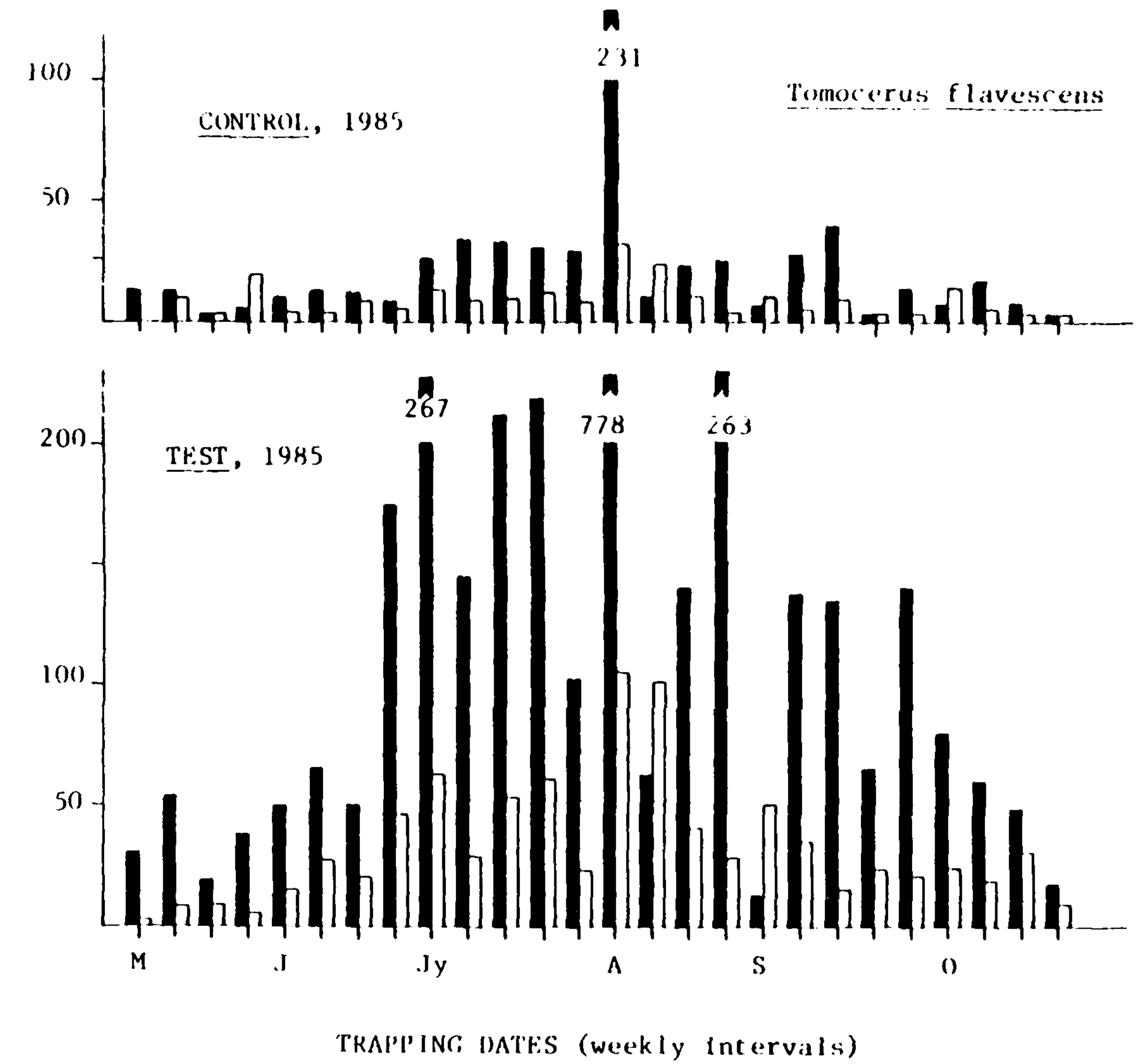


Fig. 32. Total day and night catches of Tomocerus flavescens, 1985. Open bars = day, black bars = night.

H' diversity increased in both sites, and equitability as well Bray-Curtis similarity with it (Table 15). The contribution of newly captured species, and the redistribution of dominance values, obviously affected community structure.

Table 14. Species composition and percent dominance of Carabidae, 1984-85.

SPECIES	TEST 1984	TEST 1985	CONTROL 1984	CONTROL 1985
Pterostichus melanarius Illiger	54.4	50.1	5.1	7.9
P. coracinus Newman	5.1	6.7	19.1	11.4
P. pensylvanicus Leconte	14.0	9.5	27.6	12.1
P. adstrictus Eschscholtz	1.0	0.9	15.4	10.9
P. adoxus Say	0.4	0.1	1.9	1.3
P. mutus Say	12.8	10.7	1.0	1.0
Calathus ingratus Dejean	2.7	1.0	8.6	5.7
C. gregarius Say	0.2	2.7	0.8	6.9
Calosoma frigidum Kirby	3.5	0.3	4.2	1.3
Synuchus impunctatus Say	1.8	4.8	10.3	30.4
Agonum retractum Leconte	0.8	0.8	0.5	0.6
A. decentis Say	1.4	0.8	3.2	3.2
Harpalus fuliginosus Duftschmid	0.2	3.5	0.6	2.4
Clivina fossor Linne	1.2	2.3	0.3	0.2
Cymindis cribricollis Dejean	0.4	1.2	0.5	1.5
Notiophilus aeneus Herbst	0.2	1.4	0.2	2.0
Myas cyanescens Dejean	-	0.1	1.3	0.6
Sphaeroderus lecontei Dejean	-	0.2	0.3	0.4
Agonum placidum Say	-	0.1	-	-
Trechus quadristriatus Schrank	-	0.1	-	-
Carabus sylvosus Say	-	0.1	-	0.1
Bembidion quadrimaculatum L.	-	-	-	0.1
Harpalus fulvilabris Mannerheim	-	-	-	0.1
TOTAL CARABIDAE	514	2168	623	2307
TOTAL SPECIES	16	21	18	21

ii. Barrier effects

As expected (Durkis and Reeves 1982; and our own field observations), total carabid catches were greatly augmented by barriers: by a factor of 4.2 in Test and a factor of 3.7 in Control (Table 16). Barrier factors differed between species and between sites for most species. Several which were uncommon in both sites in 1984, *C. cribricollis*, *N. aeneus* and *C. gregarius* for instance, were

Table 15. Community indices for Test and Control Carabidae, 1984-85.

	1984		1985	
	Test	Control	Test	Control
Shannon-Wiener H'	0.69	0.93	0.85	0.98
Simpson dom.	0.34	0.16	0.28	0.14
Equitability	2.94	6.45	3.57	6.75
Bray-Curtis simil.	0.35		0.42	
Sørensen's C_S	0.94		0.86	

Table 16. Barrier effects on Carabidae: increase factor for catches of species with a total N of > 10 individuals in both sites in 1985; x = N 1985/ N 1984.

	Increase x	
	Test	Control
P. melanarius	3.9	5.7
P. coracinus	5.6	2.2
P. pensylvanicus	2.9	1.7
P. adstrictus	3.8	2.6
P. mutus	3.5	4.0
C. ingratus	1.6	2.4
C. gregarius	59.0	39.5
S. impunctatus	11.4	10.9
C. cribricollis	12.5	17.5
C. frigidum	3.7	1.1
A. retractum	4.3	4.3
A. decentis	2.4	3.7
N. aeneus	31.0	46.0
N CARABIDAE	4.2	3.7

captured in much larger numbers in 1985. If year-to-year population fluctuations underlie these changes, then they occurred simultaneously in both sites (Table 16).

ii. Seasonal activity

With larger catches, we were able to document seasonal activity patterns for 14 species common to both sites. Peaks generally coincide with the animals' reproductive period (Thiele 1977), and our observations do not differ from those made by others in the US and Canada (Levesque et al., 1979, Lindroth 1969, Rivard 1964).

In Table 17, monthly captures of these 14 species are listed, and species are arranged along an early- to late- season gradient. Compared to 1984, we have added A. retractum and N. aeneus to the list of spring-breeders, H. fuliginosus to those active in early- to mid- season, and C. cribricollis to species active in mid-summer. In general, 1985 data simply confirmed 1984 observations in all cases, and th obvious synchrony of activity in Test and Control makes these data useful for future between-site comparison.

iii. Diurnal activity

In 1984, we combined Test and Control catches to assess diurnal habits of several common species. With the more ample material of 1985, we compared the behavior of 12 species between sites, and found a large number of species which would be classified as diurnal, in direct contradiction to results obtained for 1984, and to Thiele's (1977) conclusions about the nocturnal habits of forest Carabidae in general. Was increased diurnality a result of differential effects of barriers during night and day?

In Table 18, we show that increased day-activity occurred in both sites in 1985, particularly in species which were neither strictly nocturnal nor strictly diurnal in 1984 (P. coracinus, P. melanarius, S. impunctatus, C. frigidum). We also show that 1984 experiments in which barrier- and standard-traps were run simultaneously did not result in increased day catches by barriers. Therefore, we may interpret 1985 data as follows (Table 18):

Table 17. Total monthly catches of Carabidae in 1985. Major activity periods underlined. T = Test, C = Control.

		May	Ju	Jy	Au	Sep	Oct
Calosoma frigidum	T	60	6	1	-	-	-
	C	25	2	1	-	-	-
Agonum decentis	T	3	10	3	-	-	1
	C	33	31	5	1	2	1
Notiophilus aeneus	T	12	10	9	-	-	-
	C	18	11	14	3	-	-
Agonum retractum	T	7	5	4	-	-	1
	C	4	4	4	1	-	-
Harpalus fuliginosus	T	14	10	43	9	-	-
	C	15	5	25	10	-	-
Pterostichus mutus	T	92	79	21	-	7	33
	C	9	10	3	-	-	2
Pterostichus adstrictus	T	11	4	2	-	-	2
	C	110	95	29	6	7	6
Pterostichus pensylvanicus	T	75	71	26	2	9	23
	C	120	82	36	5	7	28
Calathus ingratus	T	1	1	5	7	-	8
	C	5	26	36	27	11	27
Calathus gregarius	T	4	11	26	5	2	11
	C	1	22	63	35	9	28
Pterostichus melanarius	T	17	27	448	519	75	7
	C	6	7	57	101	12	-
Pterostichus coracinus	T	5	25	50	53	11	2
	C	20	71	96	62	12	2
Synuchus impunctatus	T	-	2	60	38	3	-
	C	-	12	423	244	21	-
Cymindis cribricollis	T	-	4	11	6	1	3
	C	-	3	11	15	5	1

Strictly nocturnal species, e.g. C. cribricollis, and the single strongly diurnal species C. fossor, did not change their activity patterns through the season, but were simply caught in larger numbers during their normal period of movement.

Activity of flexible species, in response to unusually cold night temperatures in 1985, increased during the day, so that suspected "barrier factors" which were much higher for day than for night catches in these species, now became temperature factors.

Undoubtedly, year-to-year abundance variations partly determined the relative magnitude of carabid catches in 1985. However, the degree of plasticity of different species became clear in 1984-85 comparison. For full analysis and interpretation of diurnal activity, we must await 1986 data (second barrier-year, with generally warmer and more stable temperatures than in 1985). See Section IV.6. for pending statistical treatment.

Table 18. Diurnality, in % of total N, of common species of carabids in: 1984 (standard within-site trapping); 1984 barrier/ no-barrier experiment adjacent to the Control site; and 1985 (within-site barrier-trapping). (- = N < 10).

	% DIURNAL					
	1984 traps		1984 experiment		1985 traps	
Species	Test	Control	-barr.	+barr.	Test	Control
P. pensylvanicus	16.7	18.7	23.6	21.5	33.5	36.0
P. coracinus	26.9	20.7	20.0	25.9	55.5	60.5
P. melanarius	39.3	38.3	-	20.8	71.8	73.3
S. impunctatus	33.3	19.2	36.4	23.0	50.5	37.7
C. ingratus	0.0	2.9	2.9	1.7	22.7	7.6
P. adstrictus	20.0	25.7	23.5	20.0	21.1	29.8
C. cribricollis	-	-	-	0.0	0.0	0.0
C. frigidum	-	42.3	-	50.0	98.5	86.2
P. adoxus	-	0.0	-	3.6	6.7	-
A. decentis	-	-	-	0.0	0.0	2.7
P. mutus	9.1	-	-	-	19.8	16.7
C. fossor	87.5	-	-	-	89.9	-

iv. Activity of common species

Keeping in mind the above results, three species will be examined for their activity fluctuations in 1985. P. pensylvanicus (Fig. 33) remained mainly nocturnal (Table 18), and exhibited the typical spring-breeders' peak in May to late June, teneral adults of the next generation emerging in September and October.

Pterostichus coracinus, active in mid-season (Fig. 34), was more flexible in its diel responses, becoming temporarily diurnal in both sites on, for instance, August 5 and 12 (both cold nights followed by normal or warm days).

(Fig. 35)
Pterostichus melanarius, which would be considered diurnal on Thiele's scale (Thiele 1977: > 30% day-activity = diurnal) was unusually numerous in 1985 day traps. It would thus be the most plastic of the three species, and by corollary, have the lowest threshold for cold temperatures. Accordingly, its overall activity trend was the most strongly diurnal of the three in both 1984 and 1985 (Table 18).

4. Velvet mites

Of five species present in the study sites (four of them common to both), Trombidium auroraense Grandjean et al., and Abrolophus sp. were trapped with relatively high frequency. We can now distinguish between immatures of all species, and the two listed above are excellent candidates for yearly comparisons.

Both have a strictly univoltine life cycle. Abrolophus sp. (Fig. 36) probably overwinter in the egg stage, larvae appearing in early May and dispersing, presumably in search of hosts, throughout May. Deutonymphs are most active in late June, and adults through July and August, disappearing in September. In 1985, there was essentially zero overlap between life stages in both sites, and activity peaks were synchronous.

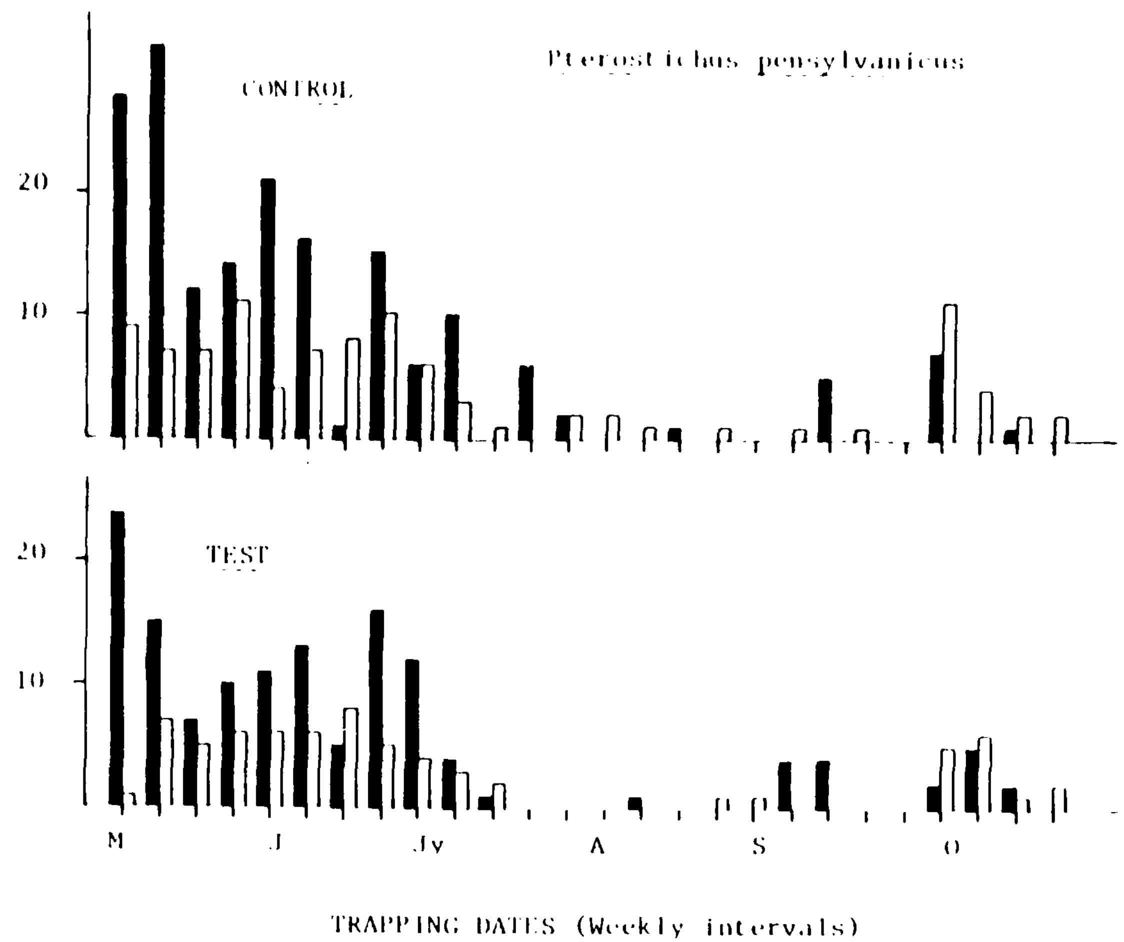


Fig. 33. Total day and night catches of P. *pensylvanicus* in 1985. Open bars = day, black bars = night.

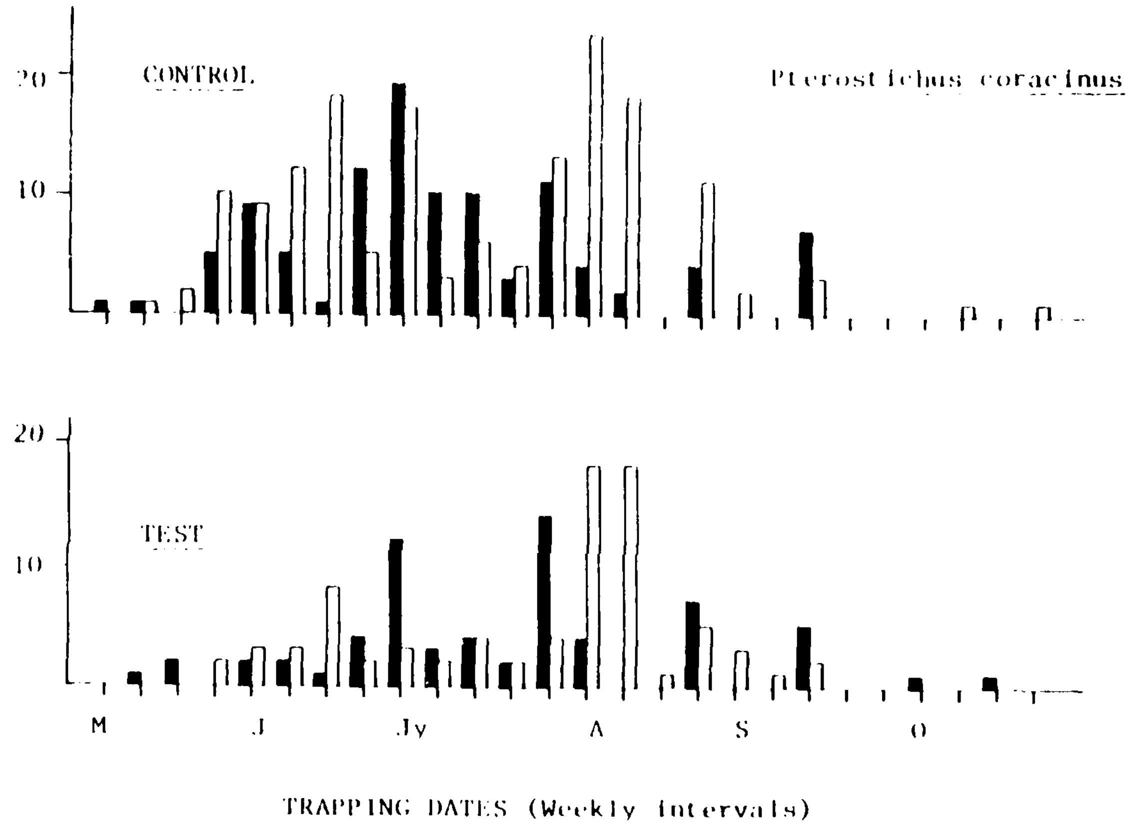


Fig. 34. Total day and night catches of *P. coracinus*, 1985. Open bars = day, black bars = night.

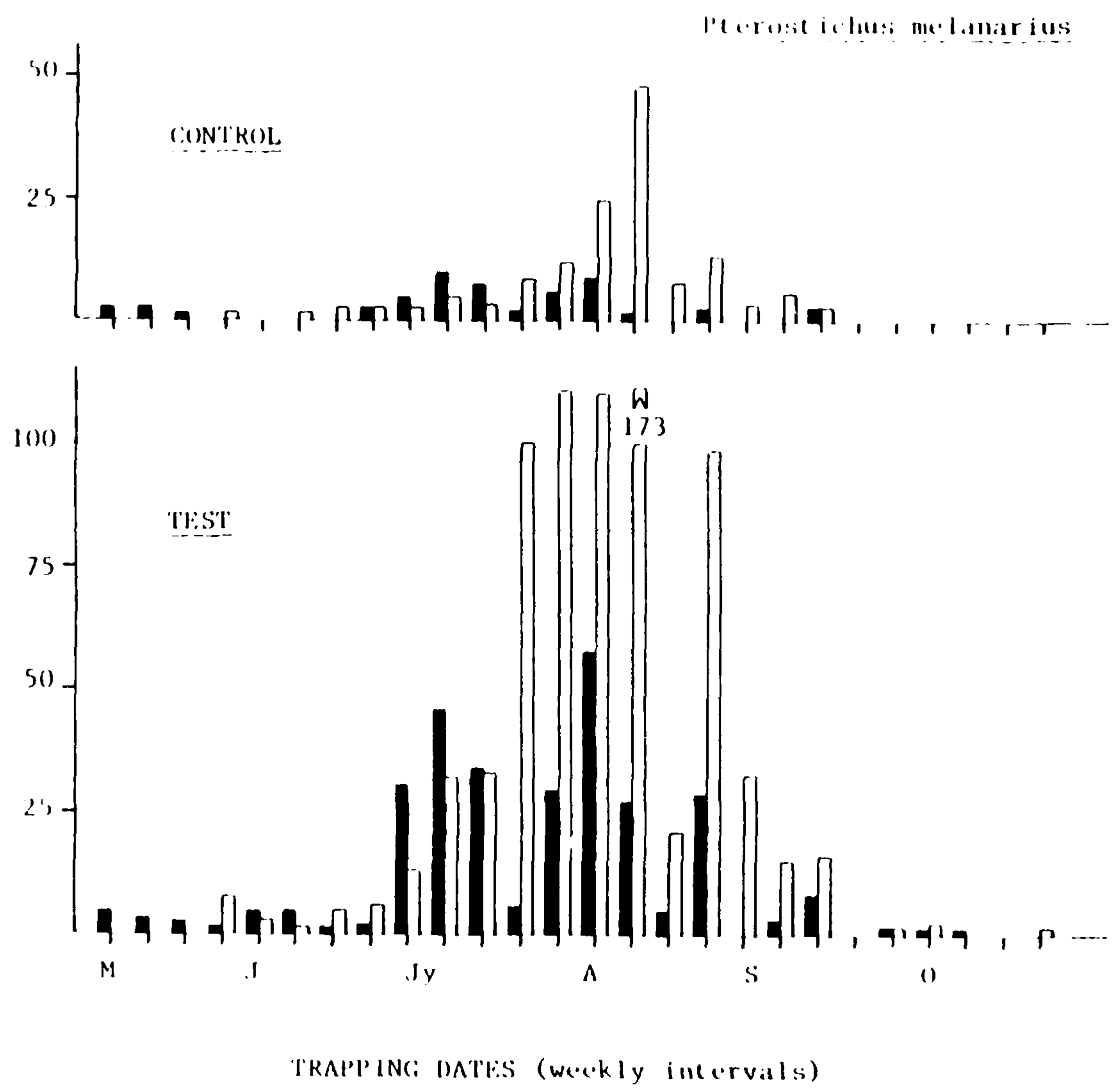


Fig. 35. Total night and day catches of P. melanarius, 1985. Open bars = day, black bars = night.

In *Trombidium auroraense*, only adults and larvae seemed to be surface-active, again with no overlap between generations (Fig. 37). Deutonymphs are occasionally recovered from soil core samples, and may not frequent the soil-surface or litter habitats. Presumably, development and maturation occur during fall; by spring, adults are active, and females lay orange egg masses in crevices of the surface soil.

Both species were mainly diurnal, and their 1985 activity patterns confirmed 1984 observations which were partly conjectural due to lower catch sizes.

5. Statistical treatment

Climatic variables are obviously the main factors impinging on overall numbers caught in traps, as well as on changes in activity patterns (nocturnal vs. diurnal). We plan to use both 1985 and 1986 data (barrier-traps) of catches of species common to both sites; regression analysis of counts on temperature and RH prior to and during trap days will test the significance of these variables, and allows testing of regression parameters between sites. Site and year affects can be tested by ANOVA (using log counts, sites, years, dates/year fixed, traps = quadrats random). Although the exact model to be used has not yet been determined, results of regression analysis will indicate appropriate climatic factors as covariates.

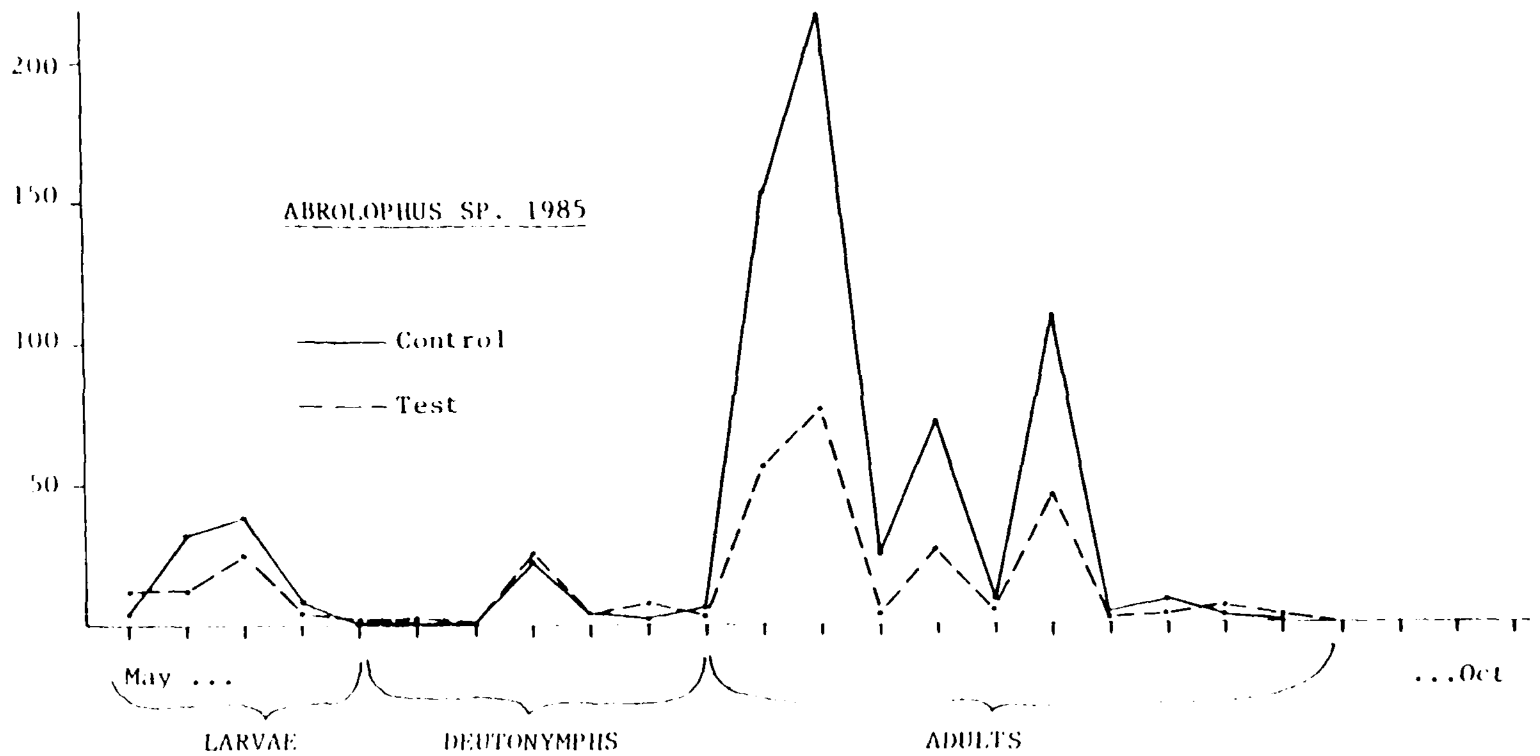


Fig. 36. Weekly catches of Abrolophus sp. in Test and Control, 1985.

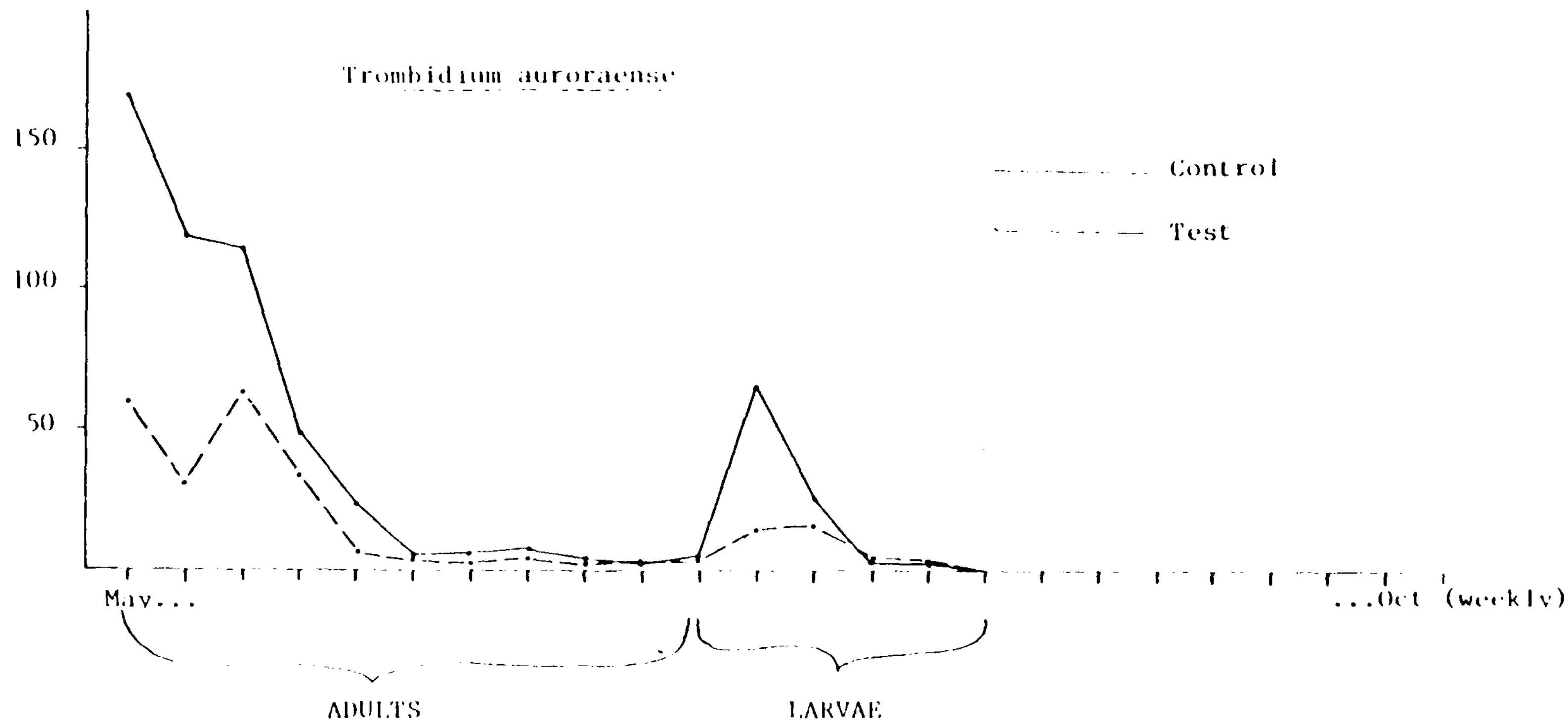


Fig. 37 .Total catches of T. *auroraense* in Test and Control, 1985.

V. LUMBRICIDAE

1. Methods and status of data

In general, methods used in 1986 were no different than those in previous years: 25 x 25 cm damples taken from litter, A and B horizons; litter was formalin-extracted, soil samples were hand-sorted and then wet-sieved (Walther and Snider 1985).

We did, however, increase sampling depth to include an additional B horizon increment (A - 30 cm), to a total depth of approximately 40 cm. Formalin applied to the bottom of A-20 cm sample holes in May of 1986 brought up a few worms with the earmarks of estivators. Beginning June 3, 1986, we have therefore made deep sampling a routine work element of the project.

Approximately 70% of 1986 samples have been identified and weighed to date. In the present report we include data for May 7 through July 14 (the last date completed for both sites). Later this spring, we intend to prepare all 1983 through 1986 data on species phenology for publication, as a single pre-ELF data package. Much detail will be left out of the present report; we focus on year-to-year density and biomass fluctuations, distribution, and seasonal patterns of weight frequency distribution.

2. Vertical distribution

i. *Dendrobaena octaedra* (Savigny)

We have shown in earlier reports (and in Snider and Snider, submitted) that *D. octaedra* behaves as a typical epigeic litter-dweller, particularly the immatures. Adults are relatively more frequent in the A horizon, where cocoons are deposited.

Although the species prefers the litter layer, its presence there is limited by dryness. A significant correlation ($P < 0.01$ for both sites) exists between % moisture of litter and % of the population present in it ($r = 0.75$ for Control,

0.80 for Test). Without doubt, the length of time litter has been moist prior to sampling, as well as temperature, may introduce variability to these data.

ii. Lumbricus rubellus Hoffmeister

This intermediate (epi-endogeic, Bouche 1977) species invades the litter layer less readily than D. octaedra, but a marginal correlation between litter moisture and % of the population found in it exists ($r = 0.62$, $P < 0.1$). The species prefers the A horizon (Snider and Snider submitted), which harbors 60-100% of the population at all times.

iii. Aporrectodea spp.

Aporrectodea tuberculata (dominant in Test) and A. turgida (dominant in Control) prefer the A horizon under favorable conditions, as evidenced by the small proportion of both populations found at lower depths in spring and fall (Figs. 38, 39). Distribution of these endogeics is clearly similar, downward migration being evident in July and August of 1984 and 1985, when A horizon moisture was below 25%. Early-season 1986 data indicate that both species retreat to depths below A-20 cm, although not to any significant extent by comparison with other Aporrectodea spp. It is likely that only a small proportion of these populations was missed prior to 1986 by not sampling a third B horizon increment.

Aporrectodea trapezoides (Control) shows a distinctly different response pattern to decreasing soil moisture, resulting in an inverted pyramid of depth distribution (Fig. 40). Recurring mid-summer droughts in 1984 and 1985 led to pronounced concentration of the species in the B horizon, to the point of leaving the A horizon entirely (late July 1985). Predictably, the unseasonable drought in the spring of 1986, which reduced A horizon moisture to < 25% in May, resulted in vertical redistribution of A. trapezoides: by mid-June, the majority of individuals were recovered from A - 30 cm (Fig. 40).

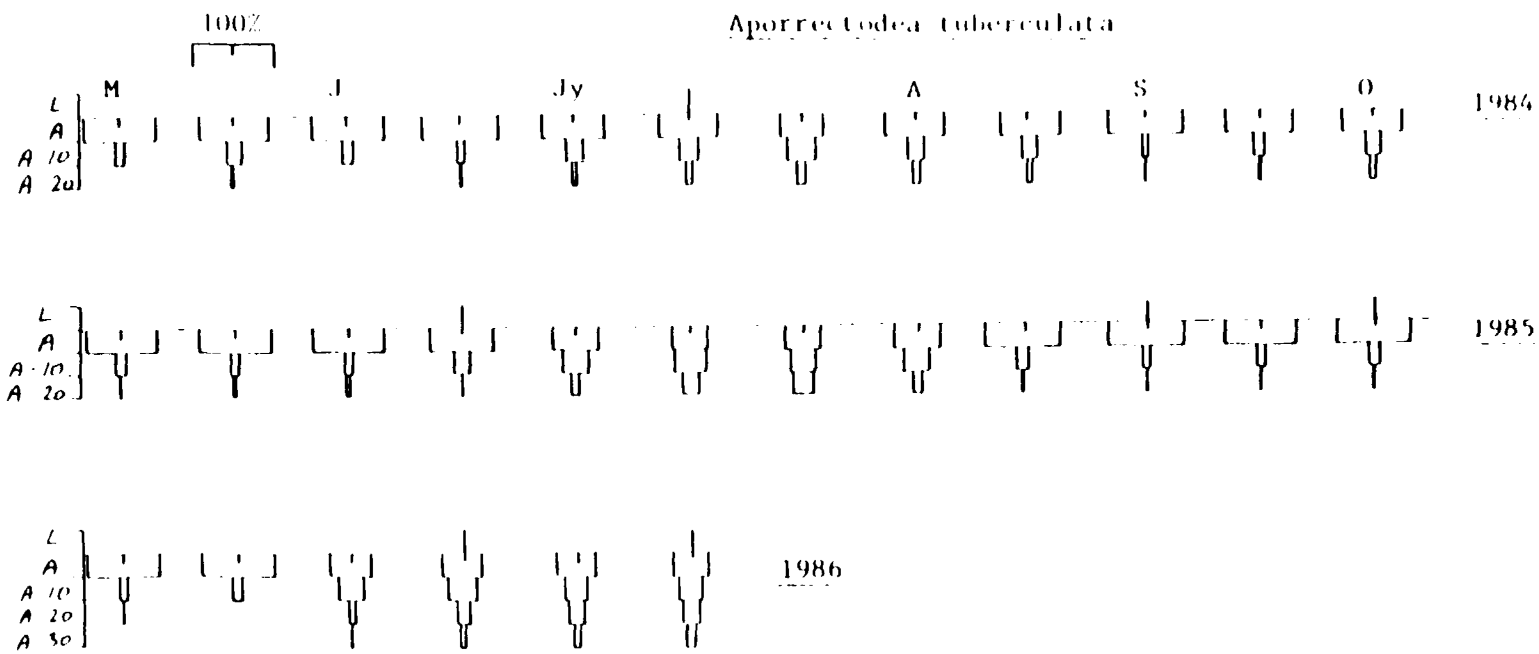


Fig. 38. Depth distribution, in % of total N/date, of A. tuberculata in Test.

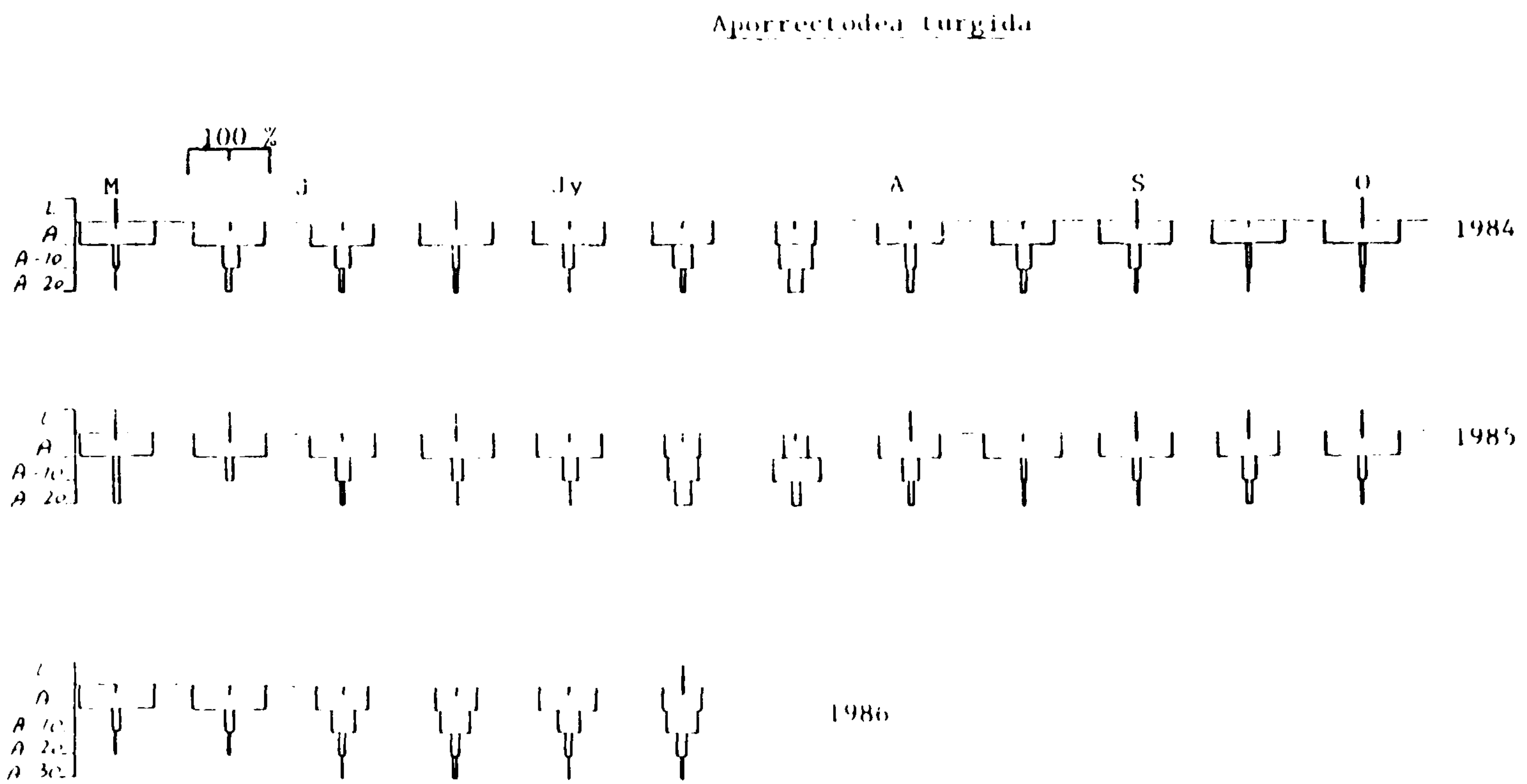


Fig. 39. Depth distribution of A. turgida in Control, in % of total N/date.

Aporrectodea trapezoides

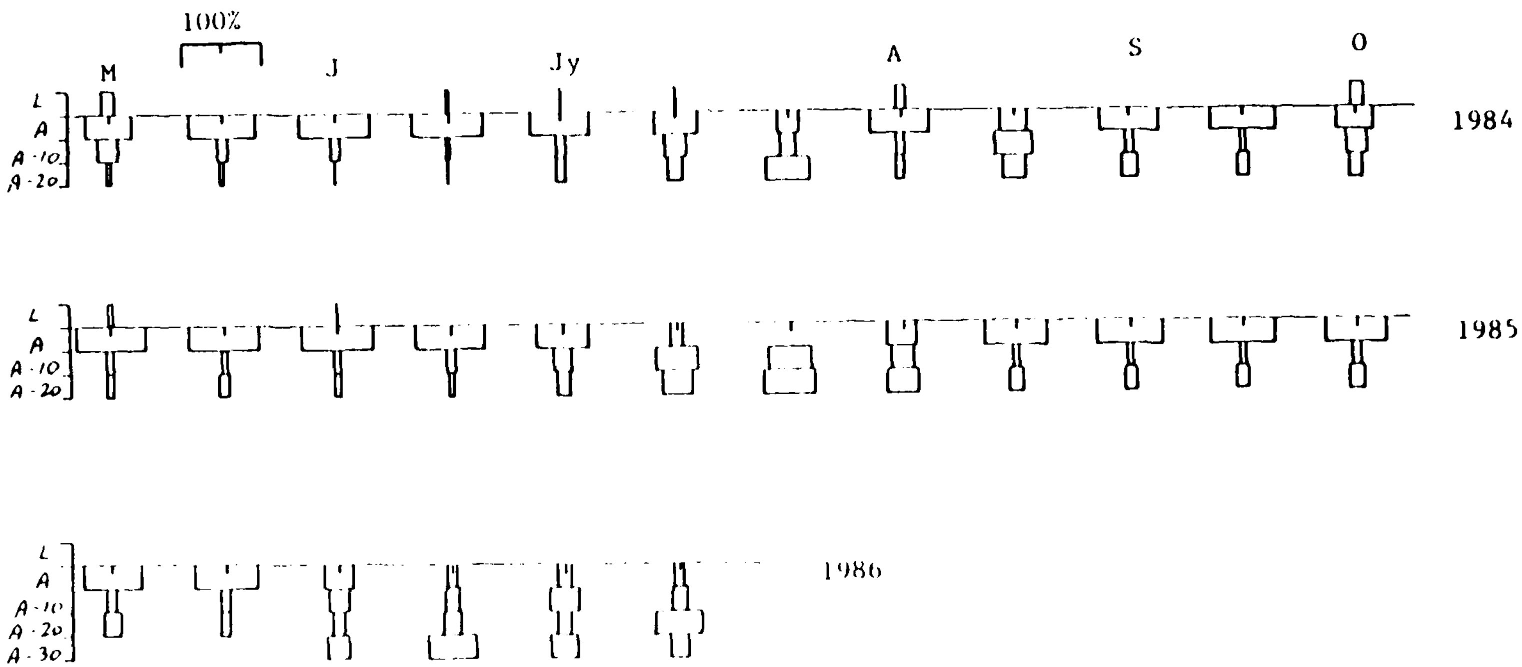


Fig. 40. Vertical distribution of A. trapezoides, in % of total N/date, in Control.

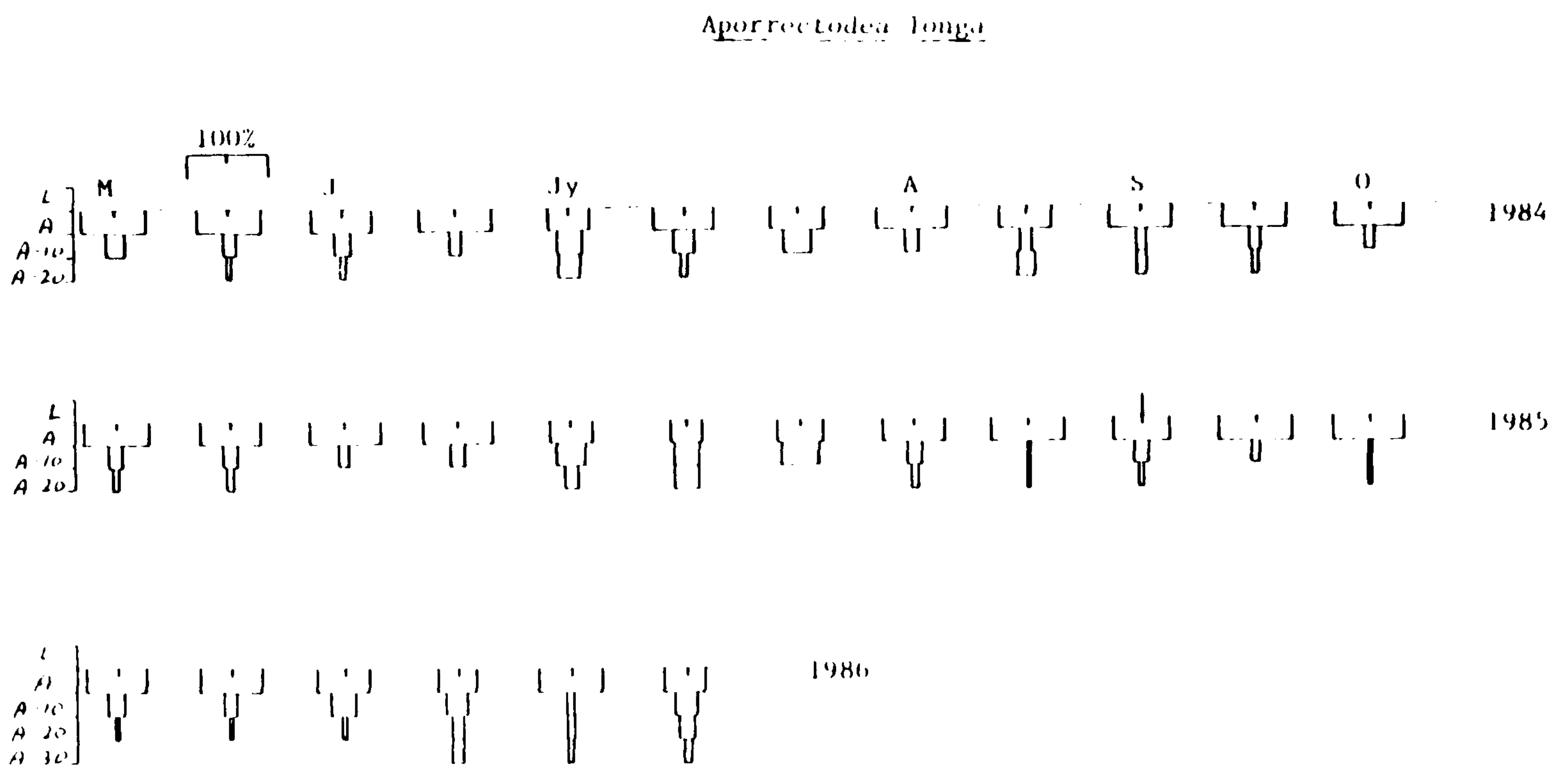


Fig. 41. Vertical distribution of A. longa, in % of total N/date, in Test.

Aporrectodea tuberculata and A. turgida thus use a strategy of slight downward migration during water stress, enduring it mainly in the A and upper B horizons. Aporrectodea trapezoides responds by pronounced vertical migration, abondoning the upper horizons almost entirely. A third strategy seems to be used by A. longa in Test (Fig. 41). Through most of the season, the species is active mainly in the A horizon, with few individuals recovered from deeper than A - 10 cm depth (e.g., May and June of 1984 and 1985). In late July of both years, when endogeics are found in the deepest horizons they are likely to occupy, A. longa disappeared from the A-20 cm increment. We speculate that this deep-burrowing form retreats to much deeper layers than we can sample, resulting in a discontinuous distribution pyramid.

For both A. longa and A. trapezoides, mid-summer population declines must be interpreted with caution, since we do not seem to catch the tail end of their vertical distribution. Unfortunately, sampling to greater depth is impossible without severe disturbance and without fragmenting most of the worms in the sample. It is preferable to continue established sampling procedures, knowing that population declines may be due to re-stratification beyond our reach rather than to mortality.

3. Horizontal distribution

Two questions were of interest with regard to earthworm distribution: a) Given that the pre-ELF period has now been extended much beyond original expectations, we were concerned about the impact of our intensive sampling regime on the ecology of the sites. All 20 quadrats within each site were sampled briefly in late 1983. Since May 1984, all even-numbered quadrats were sampled at intervals of 2 weeks, for a total of 38 sampling occasions. Since May 1985, two additional odd-numbered quadrats were sampled on a total of 26 dates. We needed to know whether we could return to sampling odd-numbered quadrats in order to

relieve the pressure placed on even quadrats, without endangering the comparability of year-to-year population estimates.

Using 1983 data (4 late-season dates, 20 samples/site), species distributions in even- vs. odd-numbered quadrats were tested in comparison with a poisson distribution. In Control, all species were distributed evenly enough ($P < 0.01$) to warrant switching sample location if necessary. In Test, A. tuberculata alone proved to be unevenly distributed ($P > 0.1$) although total numbers recovered from each set of quadrats were almost identical. We conclude that we can shift sampling locations without losing the integrity of our data, if, in Test, all 20 quadrats were sampled simultaneously on two occasions per year, in order to verify the above results.

b) Evenness of species distribution was also of interest in terms of its indicative power for potential ecological gradients over the sites. The data are being formatted for analysis; for anticipated procedures, refer to Section V. 5.

4. Density, biomass and size structure

Grouping of individuals within species populations allows us to visualize recruitment patterns and seasonal distributions of major life stages. For all species discussed below, the smallest size class encompasses individuals of approximately twice the average cocoon weight (newly emerged worms weigh about as much as a cocoon). Subsequent classes were arbitrarily chosen at 2 x increments, i.e., doubled weights from the upper limit of one class to the upper limit of the next class.

i. Dendrobaena octaedra

Despite low numbers of D. octaedra in Test, between-site comparison of population fluctuations is possible (Fig.42). Seasonal flucutations were much more pronounced in Control, but a general density increase occurred in both sites in 1985. This increase was reflected in a biomass increase in Control, less so, for this small-bodied species, in the sparse Test population (Fig. 42).

The synchronicity of cocoon densities in the two sites was more obvious than that of population abundance. Cocoon production peaked in both sites in 1984 (Fig. 43), which explains the rise in abundance in 1985; in that year, while numbers of individuals were high, cocoon production was low. As a result, populations in mid-July of 1986 did not reflect the June peaks attained in earlier years (Fig. 42). The potentially long life span of earthworms in general (Edwards and Lofty 1972) may have assured that large immatures and adults of the 1985 generation maintained a relatively high and stable biomass in 1986 (Fig. 42).

In both sites, the bulk of juvenile emergence occurred in the spring, most prominently in June (Figs. 44-45). At lower rates, juveniles continued

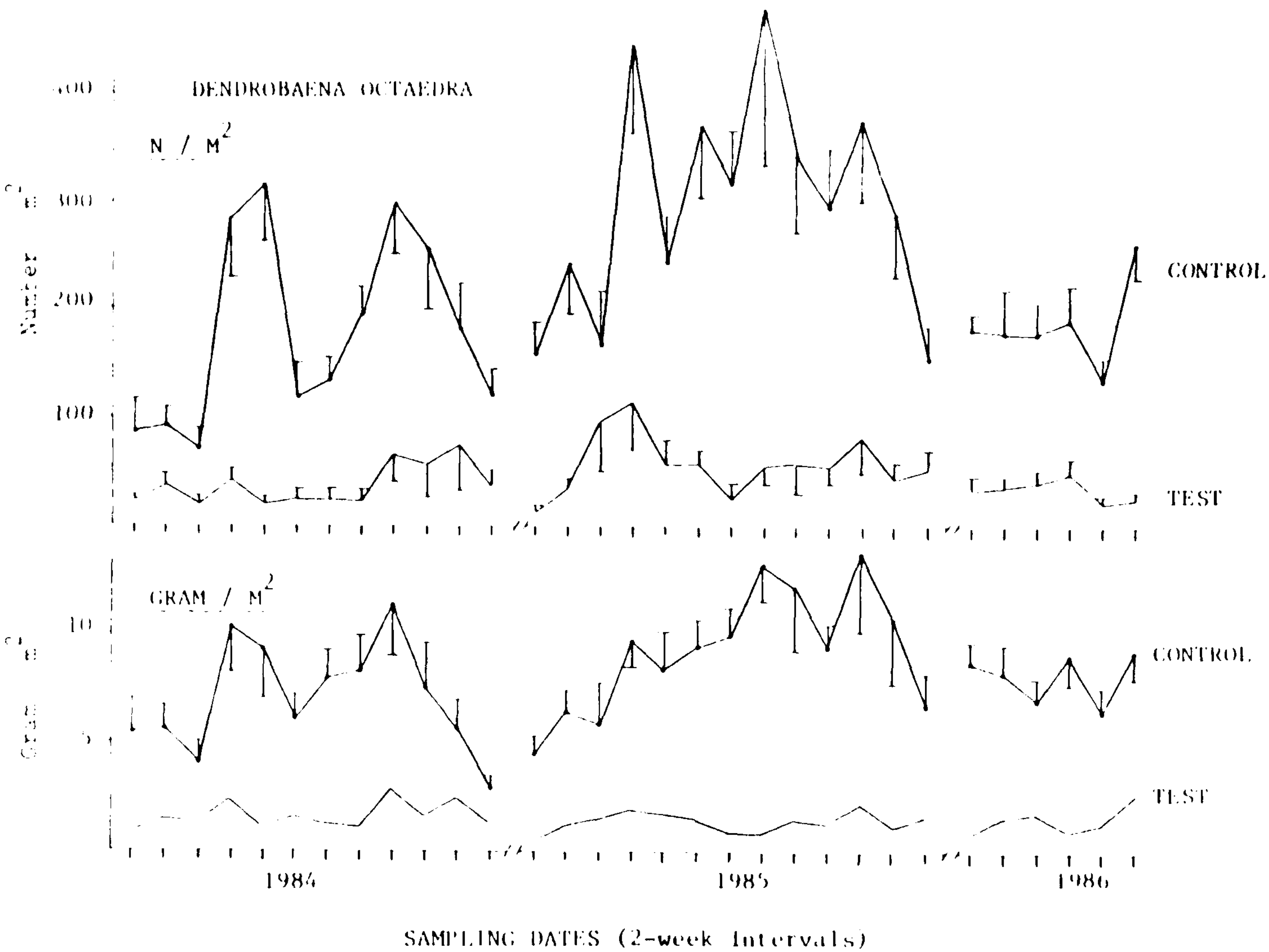


Fig. 42. Densities/m^2 ± SE of D. octaedra in Test and Control, May 1983 to July 1986; Lower graph: biomass/m^2 ± SE.

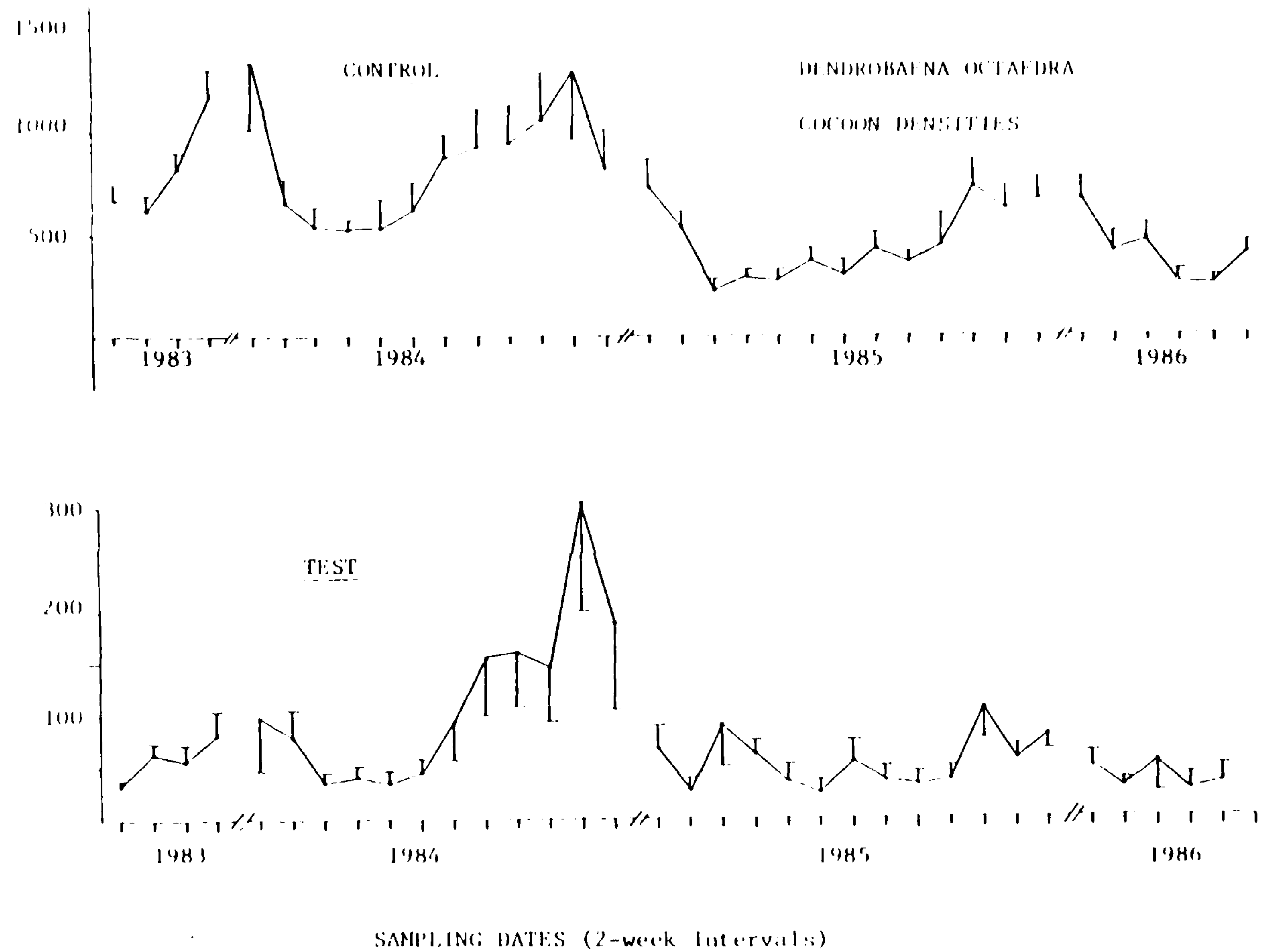


Fig. 43. Cocoon densities for D. octaedra ± SE, August 1983 to July 1986.

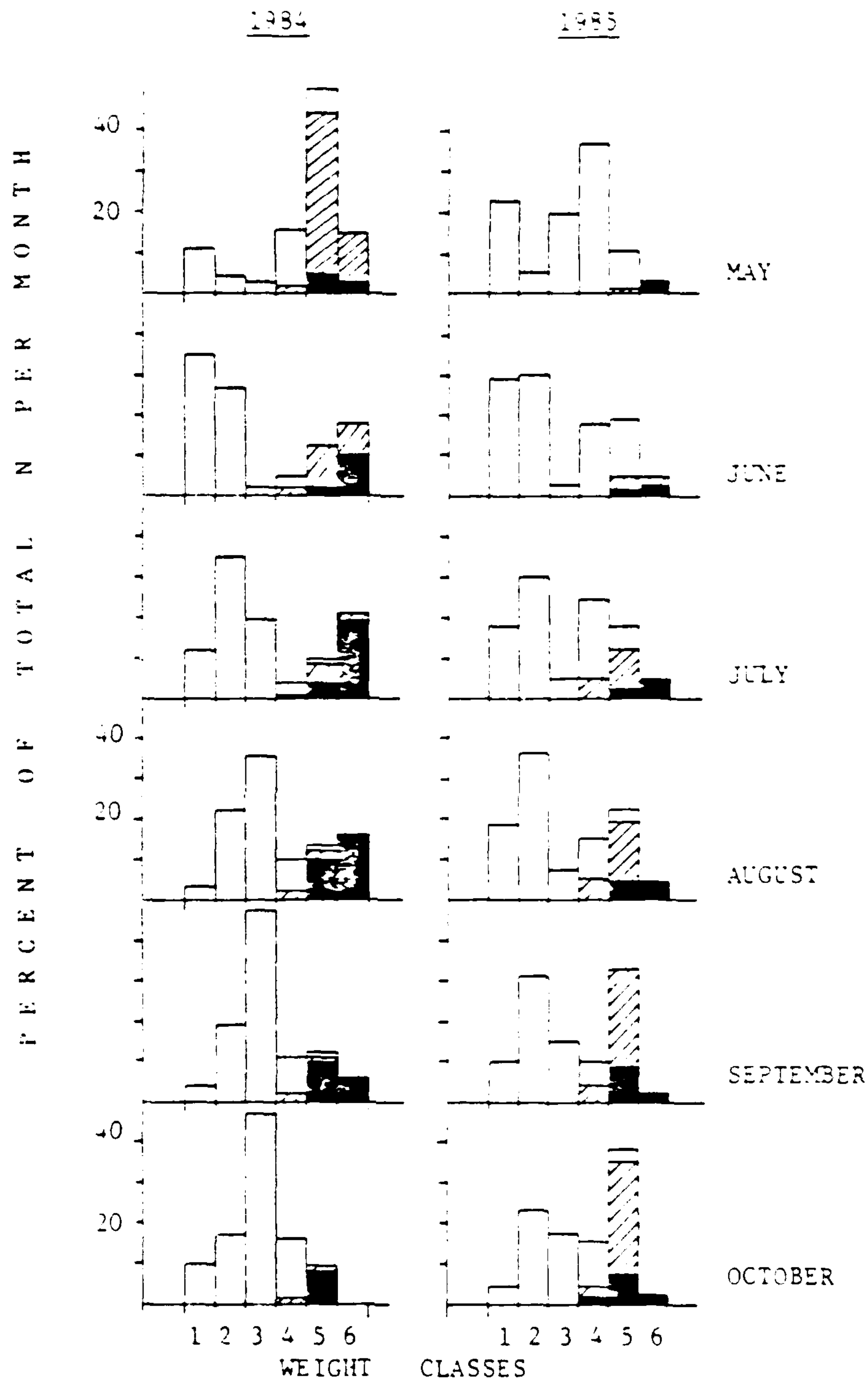


Fig. 44. Size class structure of D. *octaedra* in CONTROL, 1984 and 1985. Class 1: ≤ 0.006 g; class 2: 0.0061 - 0.012 g;.... class 6: > 0.96 g. Black bars: clitellate adults; hatched bars: aclitellate adults.

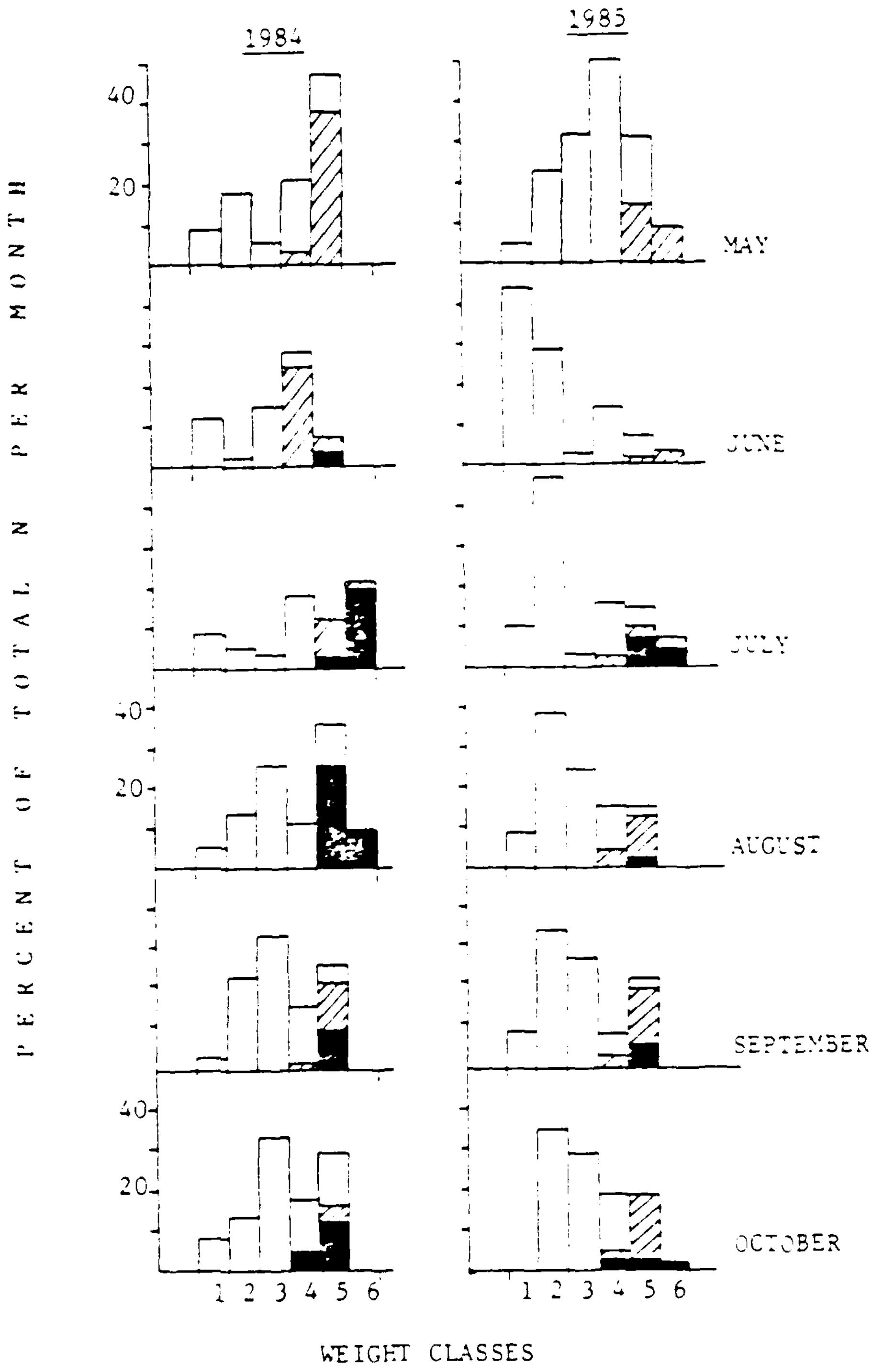


Fig.45 . Weight class distribution of D. octaedra in TEST, 1984-85.

For explanation of classes and symbols see Fig. 47.

to enter the population through summer and fall. Of particular importance was the difference between years in terms of reproductive activity and cocoon densities (Fig. 43). In Control, cocoon densities in late 1983 - early 1984 were high (mainly cocoons in advanced stages of development) and by June of 1984, 35% of the population consisted of small immatures (Fig. 44). A large percentage of clitellate adults were present throughout the summer of 1984, cocoon densities were considerable, and recruitment from May to August of 1985 was correspondingly elevated (Fig. 44). During 1985, however, clitellates were less frequent in the population, cocoon production was depressed, and size class 1 juveniles made up less than 10% of the population in May and June of 1986 (data not included in Fig. 44).

In Test, the pattern was essentially the same. Cocoon abundances of one year furnished a good predictor for recruitment rates the following season: cocoons in Test in late 1983 were relatively less abundant than in Control; consequently, the smallest size class in spring of 1984 was less well represented than in Control (Fig. 45).

ii. Lumbricus rubellus

Year-to-year variation in abundance was pronounced in this species (Fig. 46). Numbers were stable through 1984, but increased in 1985 through addition of immatures hatching from cocoons present in the fall of 1984. Both abundance and cocoon densities were variable in 1985. However, there was again a good correlation between reproductive parameters and population size structure.

Increased cocoon densities in 1984 (Fig. 46) corresponded to high frequencies of clitellates, and resulted in a relatively prolonged emergence period in 1985 (Fig. 47). Small immatures generally were most frequent in the spring, hatching from overwintered cocoons, although some of the

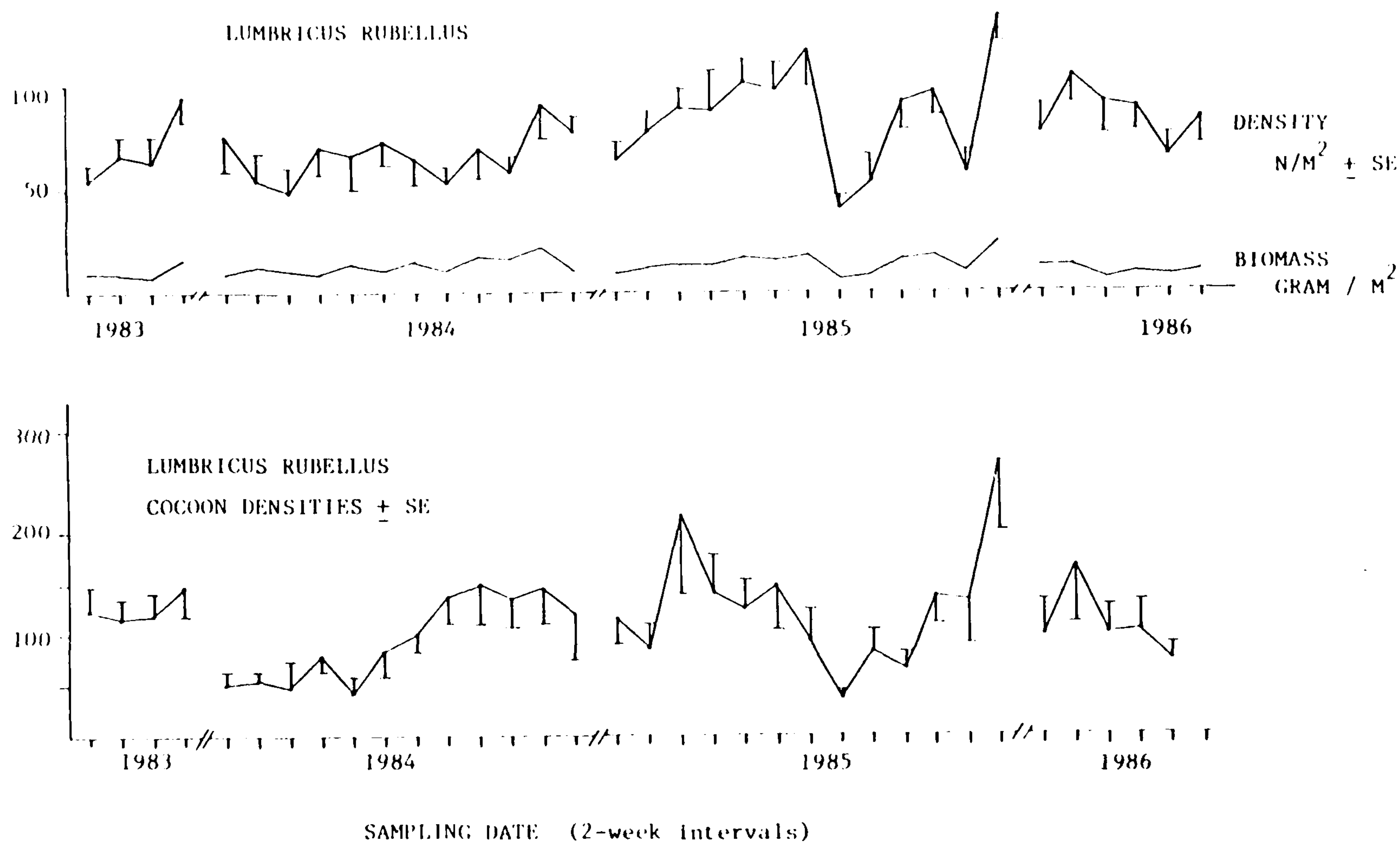


Fig. 46. Density and biomass of L. rubellus in Test, August 1983 to July 1986 (upper graph); cocoon densities/m^2 for the same period (lower graph).

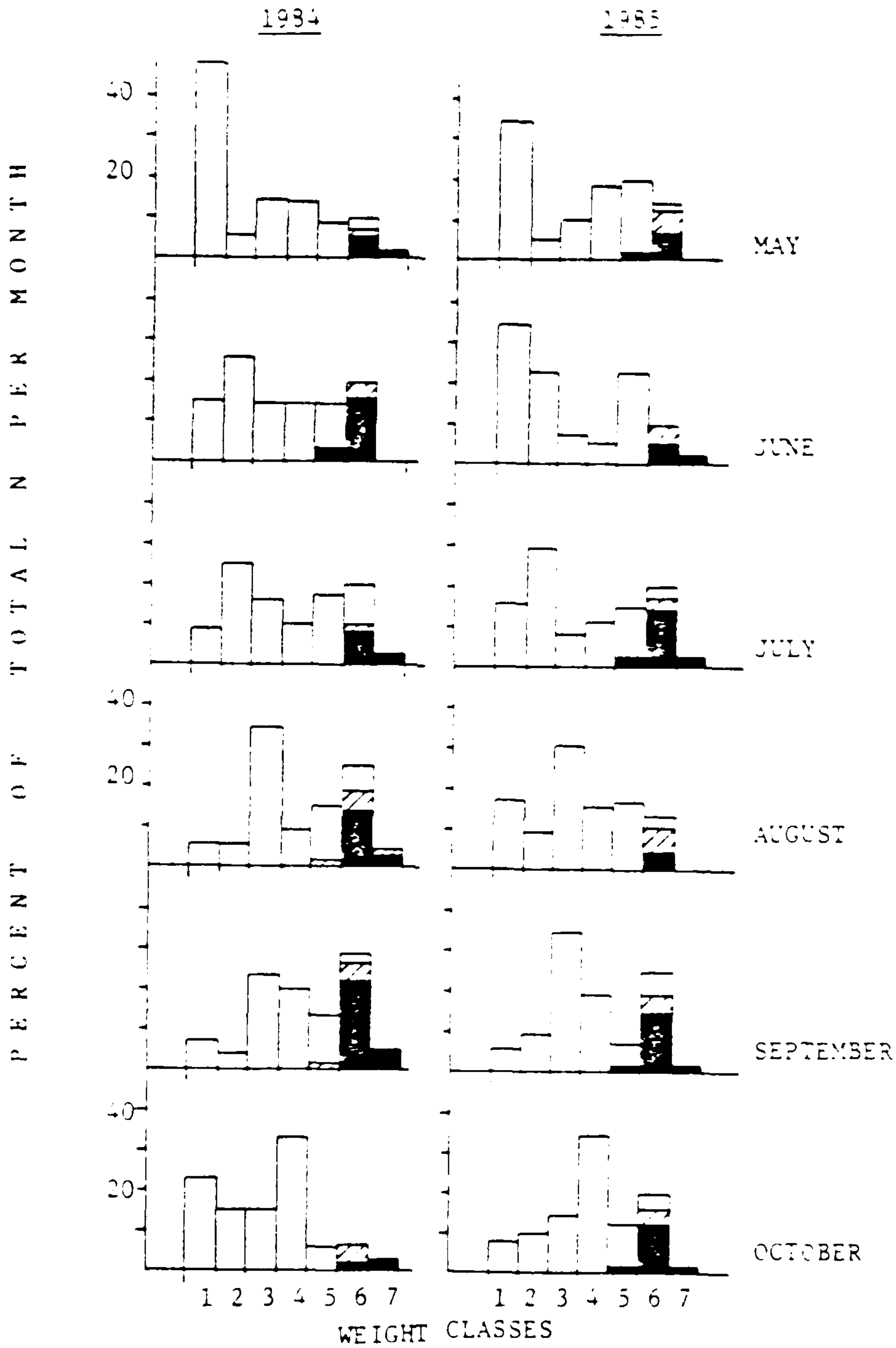


Fig. 47. Weight class distribution for L. rubellus, in TEST, for 1984 and 1985. Class 1: ≤ 0.02; class 2: 0.021- 0.04;..... class 7: > 0.640 g.

cocoons laid during spring and summer matured and hatched in the fall of the same year (e.g., October 1984, Fig. 47).

iii. Aporrectodea spp.

By European nomenclature, A. tuberculata and A. turgida would be conspecific (Reynolds 1977). In our sites, they showed essentially identical vertical distribution patterns and responses to moisture stress (Figs. 38-39). Density estimates, however, were similar only in that mid-summer declines occurred in both species in 1985 (Fig. 48). A more powerful tool of species comparison was provided by estimated cocoon densities (Fig. 49). Both species increased cocoon production in the fall of 1984; these cocoons overwintered and gradually hatched during the first half of 1985. The seasonal reproductive pattern of 1984 was not repeated in 1985, and cocoon densities in early 1985 were accordingly low for both species (Fig. 49).

Emergence times were clearly synchronous for A. turgida and A. tuberculata, and exemplify the general lumbricid adaptation to changing environmental conditions: cocoons may be produced at any time of the season, i.e., there is no set reproductive period, resulting in emergence patterns which can vary markedly from year to year (maxima in August-September 1983; spring-summer 1984; and July-September 1985, Figs. 50-51).

Abundance estimates of A. trapezoides, which occurs only in the Control site, reflected moisture conditions rather than true densities, since the species retreats to depths not sampled prior to 1986. Thus mid-summer declines in 1984 and 1985 (Fig. 52) simply indicated vertical displacement. However, the data indicated more clearly than in other species that cocoon development in our climate takes a full year. Cocoons were most abundant in August of 1984

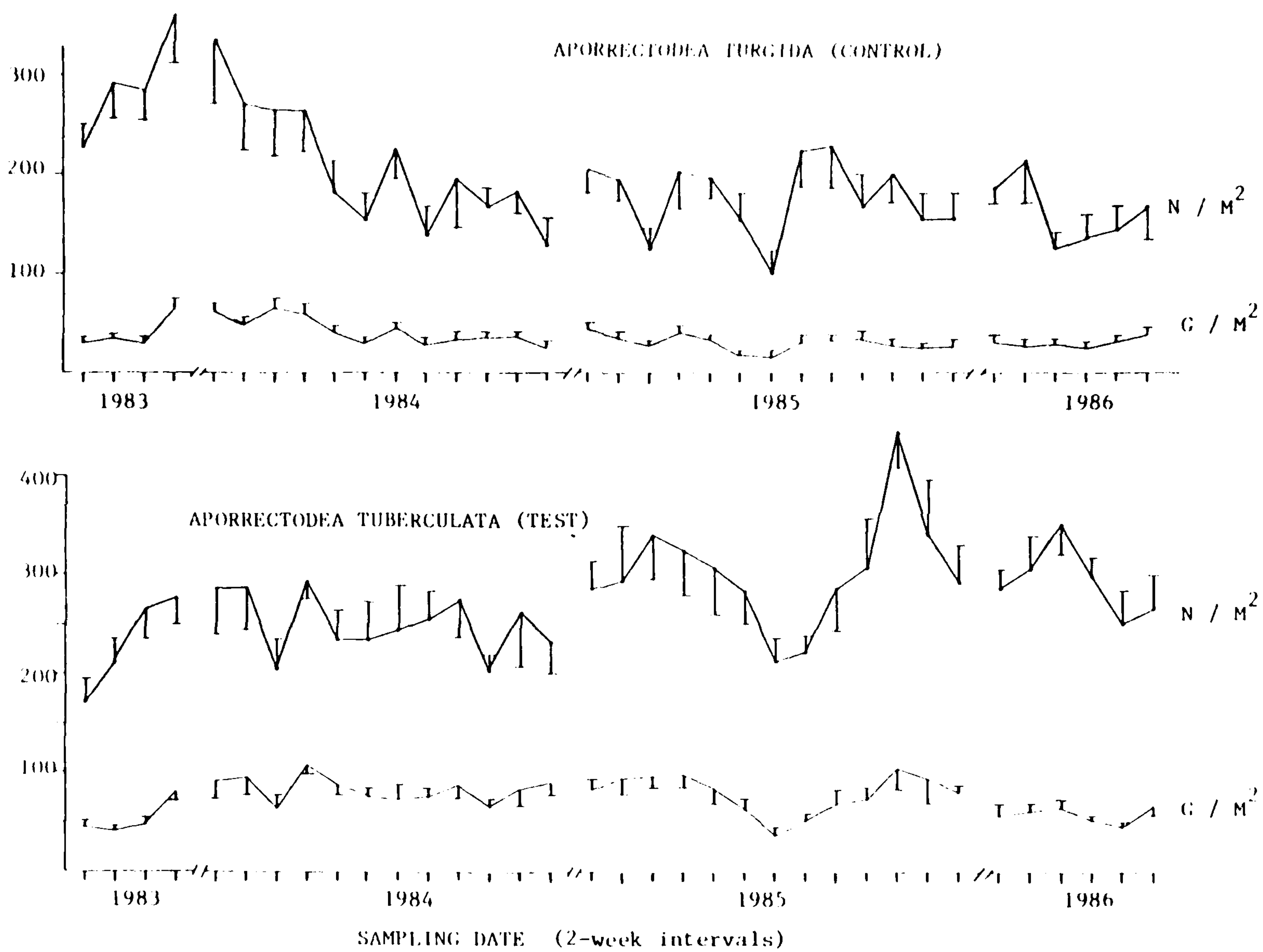


Fig. 48. Densities and biomass/ m^2 $\pm$SE for *A. tuberculata* in Test and *A. turgida* in Control, August 1983 to July 1986.

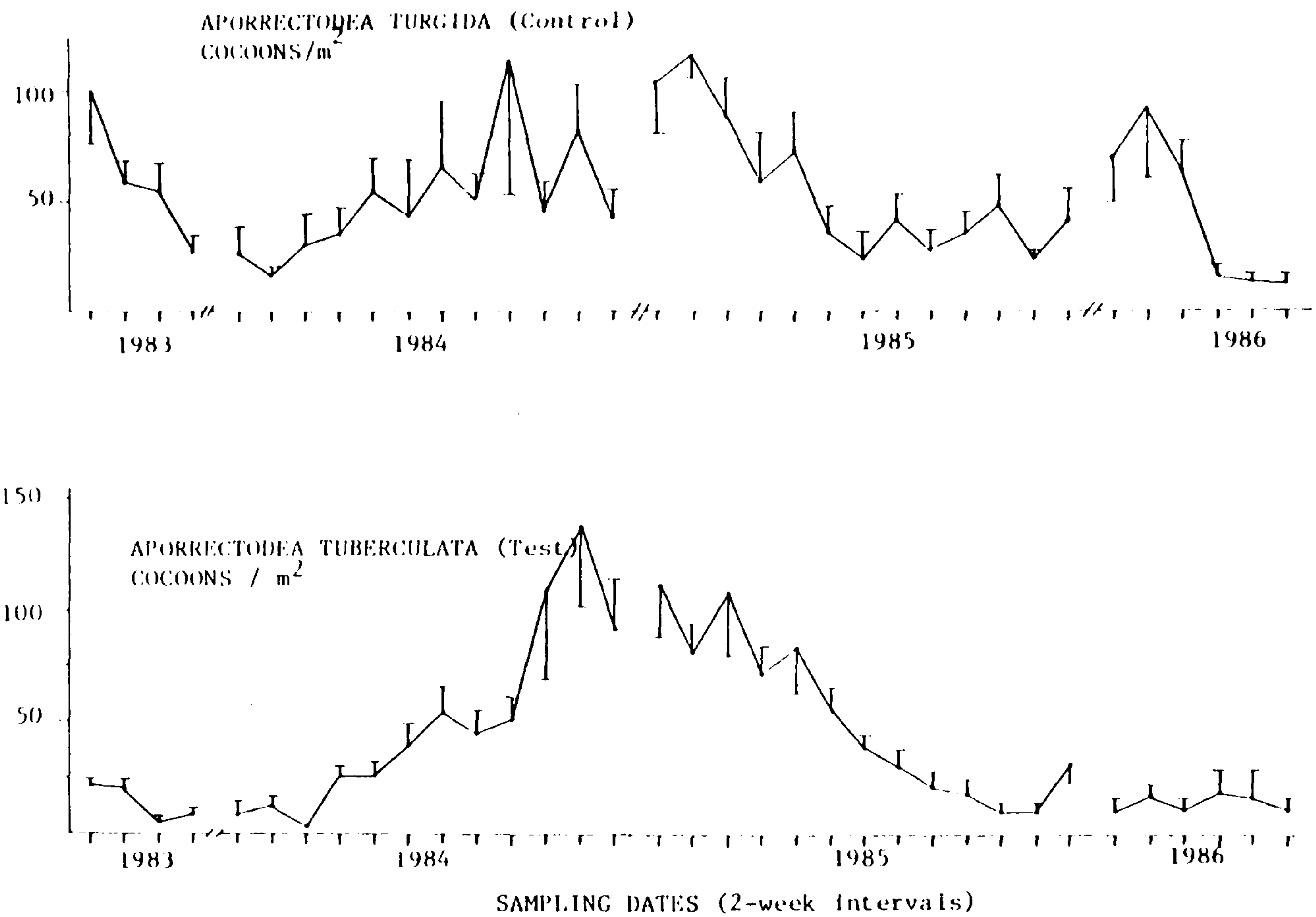


Fig. 49. Coccon densities ± SE for A. turgida and A. tuberculata, August 1983 to July 1986.

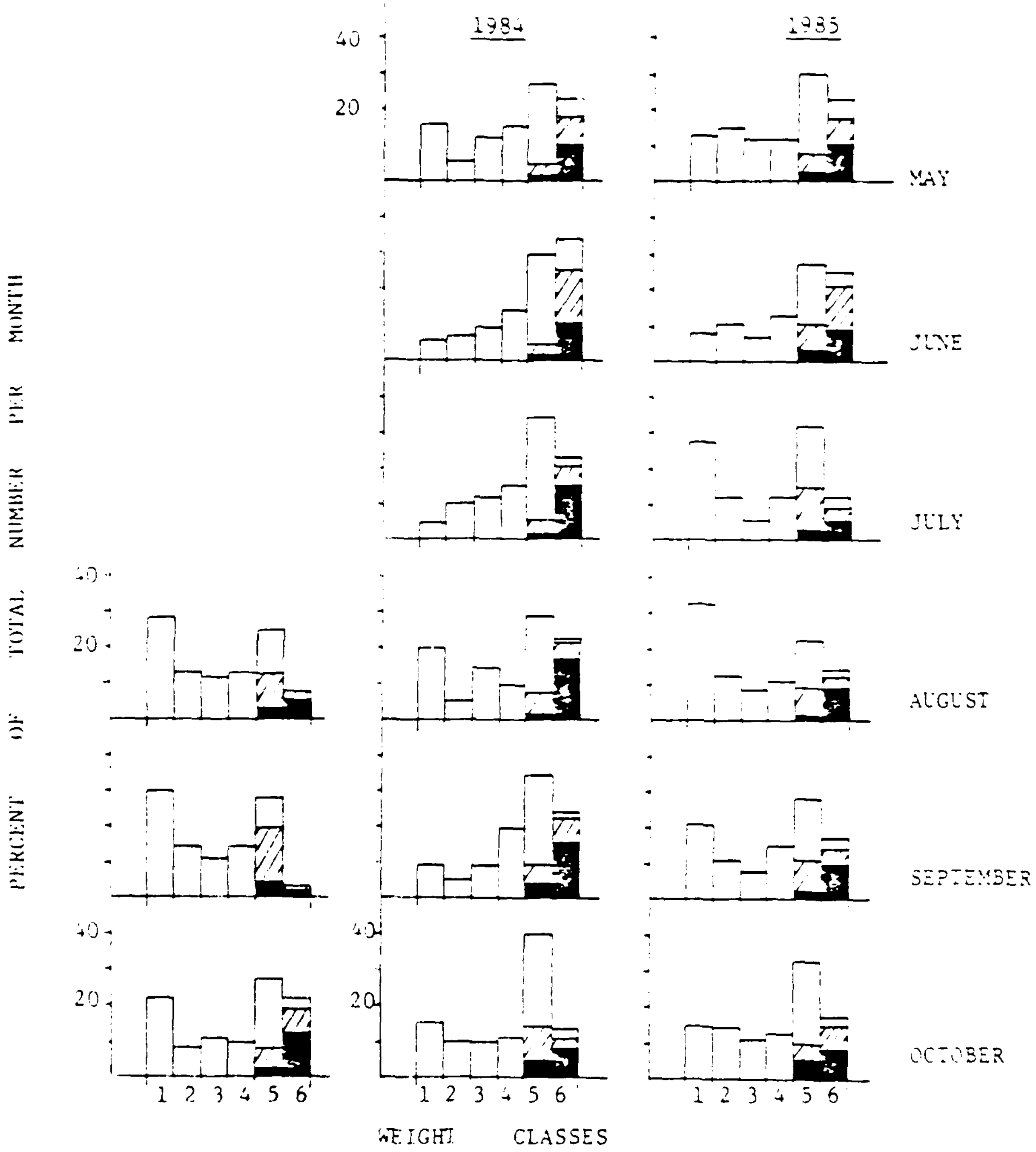


Fig. 50. Weight class distribution of *A. turgida* in CONTROL, August 1983 to October 1985. Black bars = clitellates; hatched bars = aclitellates; open bars = immatures. Weight class 1: ≤ 0.02 g; class 2: 0.021-0.04 g.... class 6: > 0.320 g.

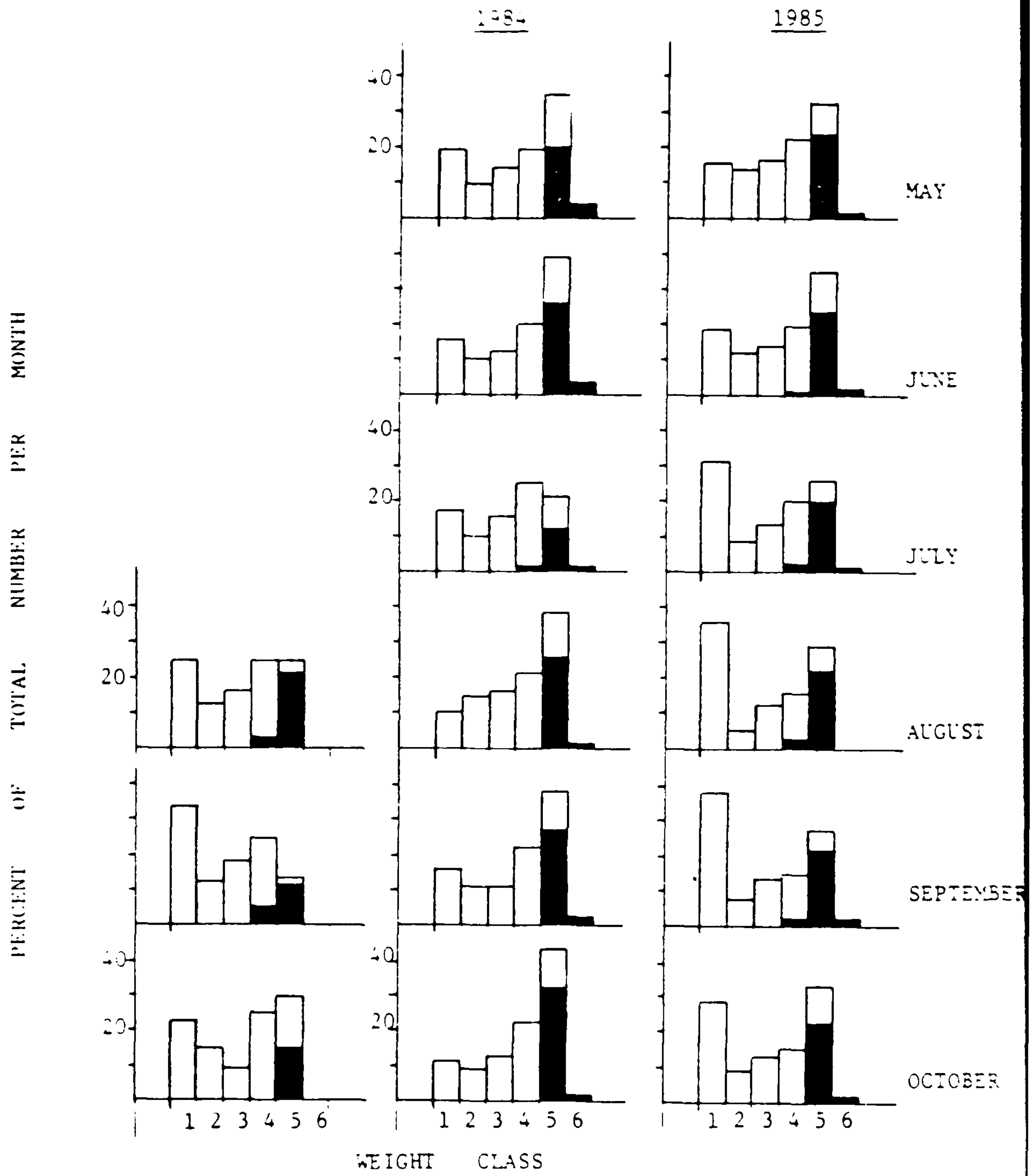


Fig. 51. Weight class distribution of A. tuberculata (TEST), August 1983 to October 1985. Black bars = all adult developmental stages; Class 1: ≤ 0.05 g; class 2: 0.051-0.10 g;.... class 6: > 0.800 g.

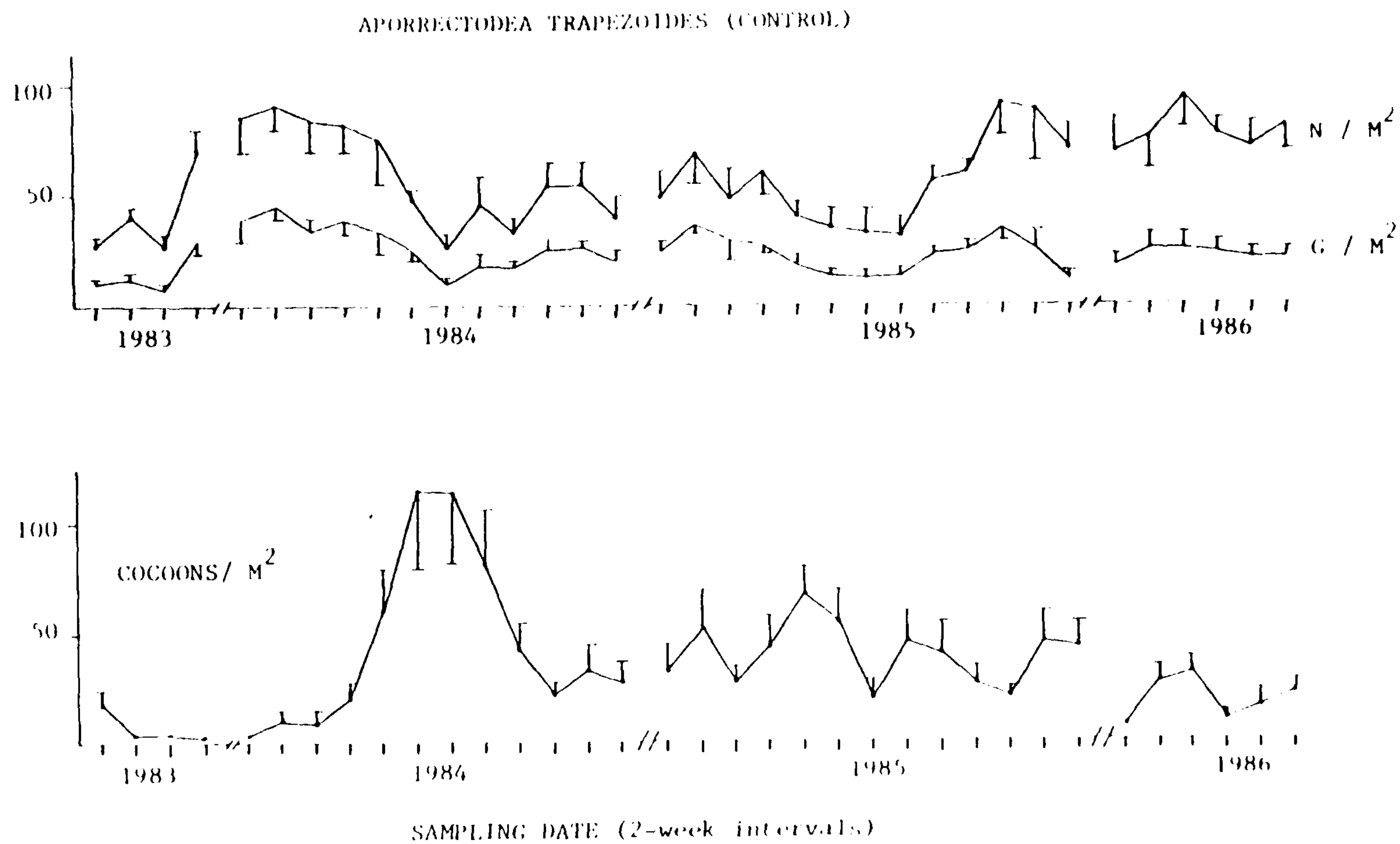


Fig. 52. Density and biomass/m^2 $\pm$ SE for A. *trapezoides* (Control), August 1983 to July 1986. Lower graph: cocoon densities / m^2 $\pm$ SE.

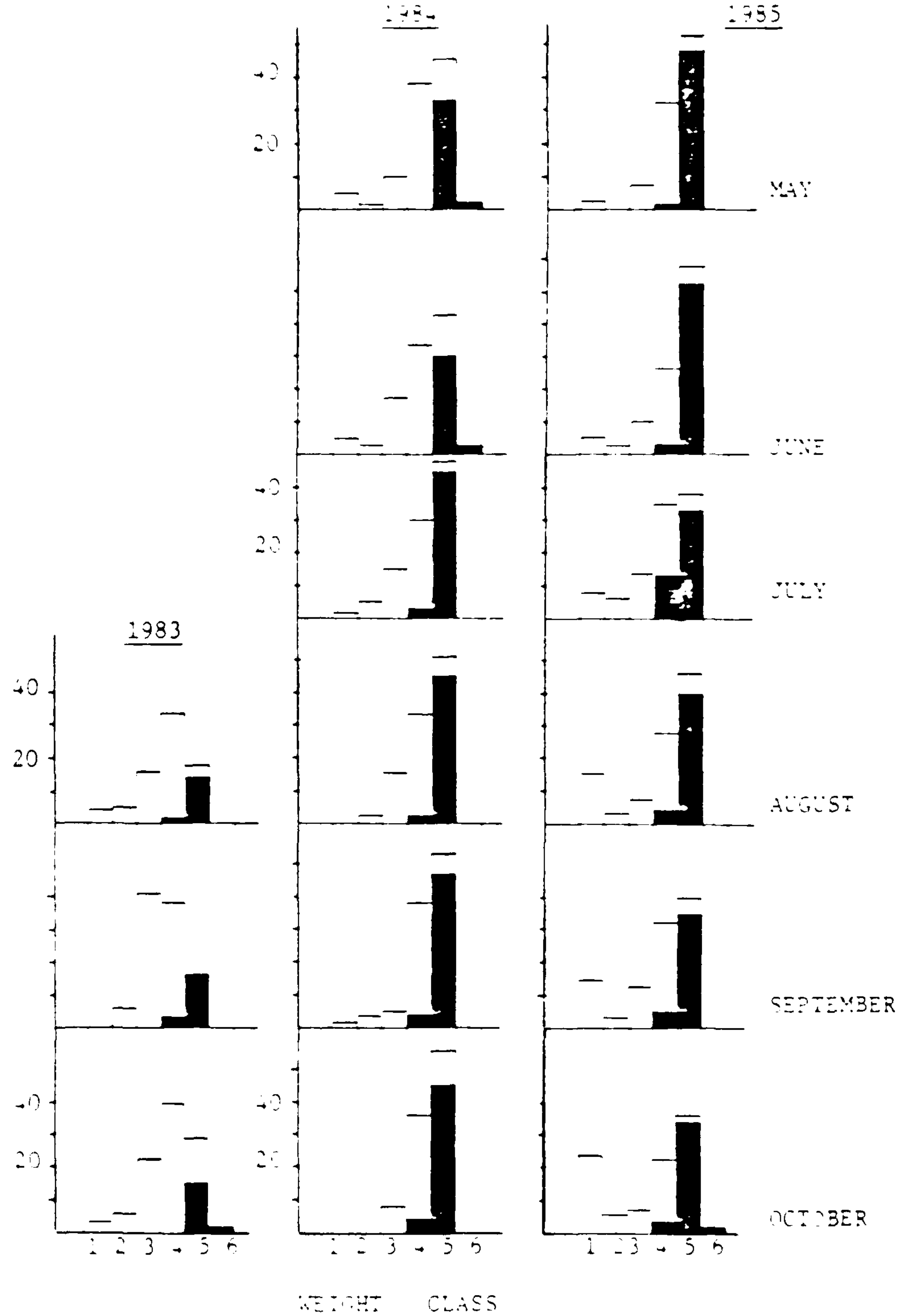


Fig. 53. Weight class distribution for A. *trapezoides* (Control), August 1983 to October 1985. Black bars = all adults; open bars = immatures; class 1: ≤ 0.05 g; class 2: 0.051-0.17 g;...class 6: > [illegible].8 [illegible] g.

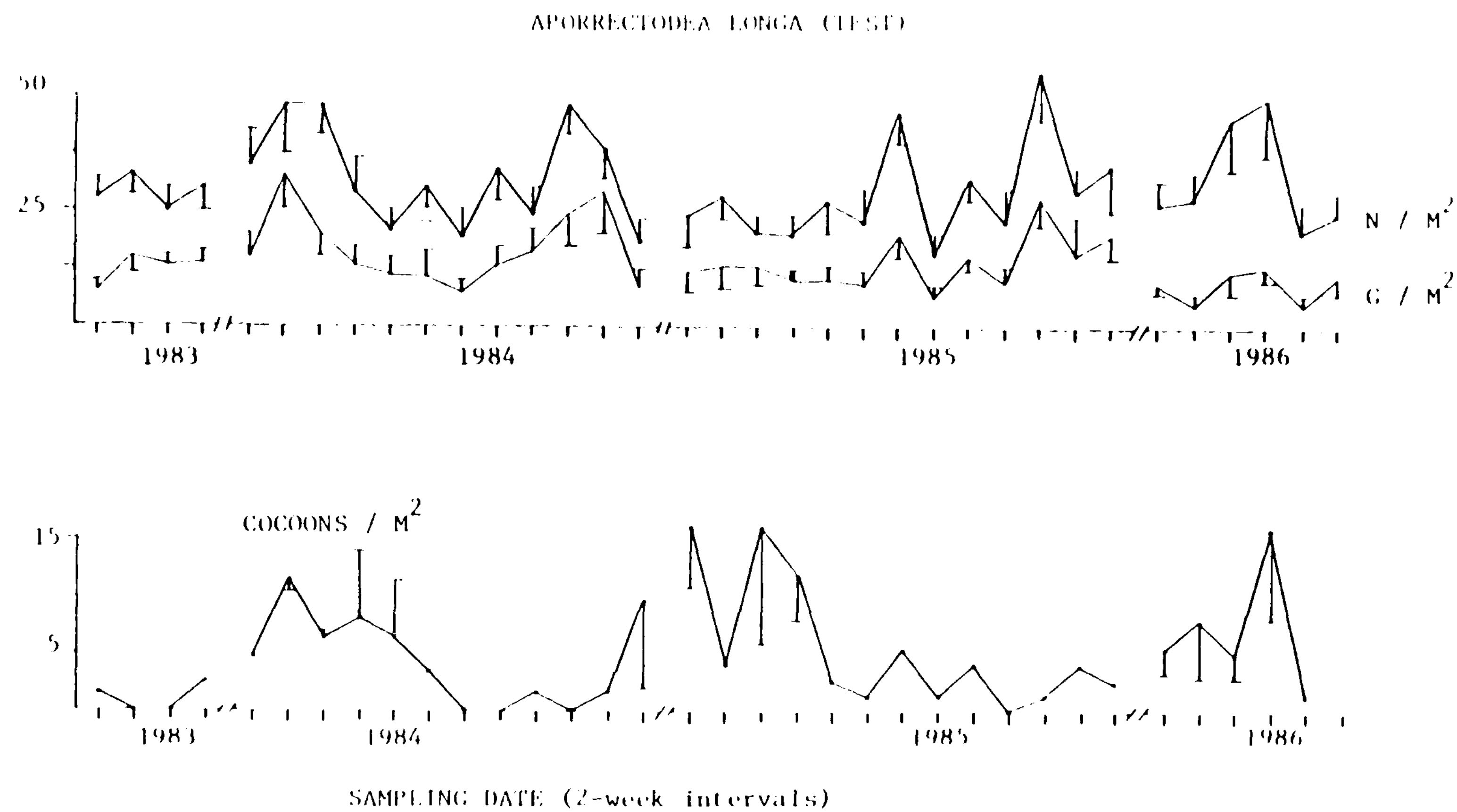


Fig. 54 . Density and biomass/ m^2 $\pm$ SE of A. longa in Test, August 1983 to July 1986.

Lower graph: cocoon densities/ m^2 $\pm$SE.

(Fig. 52), yet small immatures did not appear in appreciable numbers until summer of the following year (Fig. 53). Conversely, virtual absence of cocoons in 1983 (Fig. 52) corresponded to absence of small juveniles in the second half of 1984 (Fig. 53).

Population fluctuations of A. longa, an anecic species occurring only in Test, must be interpreted with as much caution as those of A. trapezoides, and for the same reasons. Deep vertical migration during the summer weeks (Fig. 41), possibly accompanied by obligatory diapause (Bouche 1972), appeared as spring and fall maxima in our 1984 data (Fig. 54). Characteristic of anecics (Bouche 1977), A. longa produced very few cocoons, and total numbers of both worms and cocoons recovered from samples were usually low. We expect that 1986 data, stemming from samples taken to greater depth, will elucidate the life cycle of A. longa.

5. Statistical treatment

Analysis of horizontal distribution of earthworms is currently being prepared for. We intend to use multivariate ANOVA (quadrats random, dates fixed), each cell containing an array of $y_{1...5}$ counts of individuals in 1...5 species. We will use an appropriate transformation to normalize the data ($y' = \log_e (y+1)$). Results will indicate whether species arrays (= community composition) differ significantly with quadrat (= over the site as a whole).

Density fluctuations by themselves are probably not very sensitive measures of the state of a population of earthworms. What is more important is the structure of populations in terms of numbers/weight class, and in terms of the frequency of reproductive/non-reproductive individuals in a given class.

We anticipate analysis of frequency distributions by means of contingency tables. Examples, using arbitrary numbers, are given below.

a) for any one species, year-to-year variation can be tested by, for instance, a 24x6 table with three classifications: A = year 1, 2; B = dates 1 to 12 within years; C = weight classes 1 to 6. Each cell contains N observations / class / date. Useful summary statistics include: 2x6 tables (2 years, 6 classes), with observations summed over B (dates) to test overall yearly differences; 12x6 tables (12 dates, 6 classes) with observations summed over A (years), to test overall seasonal shifts in frequencies as they may characterize each species.

b) For taxa shared between sites (D. octaedra, and the ecologically similar species pair A. turgida and A. tuberculata), tables including an additional site classification for Test and Control test differences between sites. With several years of pre-ELF data at hand, observations can be summed over years and [2 (sites) x N (years)] tables will eventually test whether significant overall changes take place following antenna operation.

VI. LITTER INPUTS AND DECOMPOSITION

1. Review

Earlier hopes of documenting decomposition as well as nutrient loss rates cannot be fulfilled. Several constraints prevent us from pursuing nutrient analyses further. Primarily, available manpower, which was originally calculated for faunal analyses only, is far too limited for gathering and processing nutrient samples with the degree of accuracy needed for valid results. Rather than obtain insensitive nutrient data, we opted to invest manhours into routine validation of faunal extractions, and to continue barrier-trapping (which increases sorting and identification time). We believe that the decomposition work elements we did chose to continue will yield a restricted, but useful data base for pre- and post-ELF comparison.

Specifically, we retain monitoring of litter inputs and standing crops, the latter with some changes in technique; and monitoring of decomposition rates using the leafpack technique. Litterbag studies, which have proven unsatisfactory with respect to obtaining realistic decomposition rates (1985 annual report) will be discontinued, as will nutrient analyses of litter samples.

2. Litter inputs

The temporal pattern of abscission in 1986 resembled that observed in 1984, the bulk of maple leaves falling within a period of approximately 1.5 weeks (Fig. 55). As in previous years, leaf fall in Test and Control was tightly synchronized. Analysis of variance of 1984 through 1986 data showed that neither site and year effects, nor their interaction, were significant ($P \gg 0.1$).

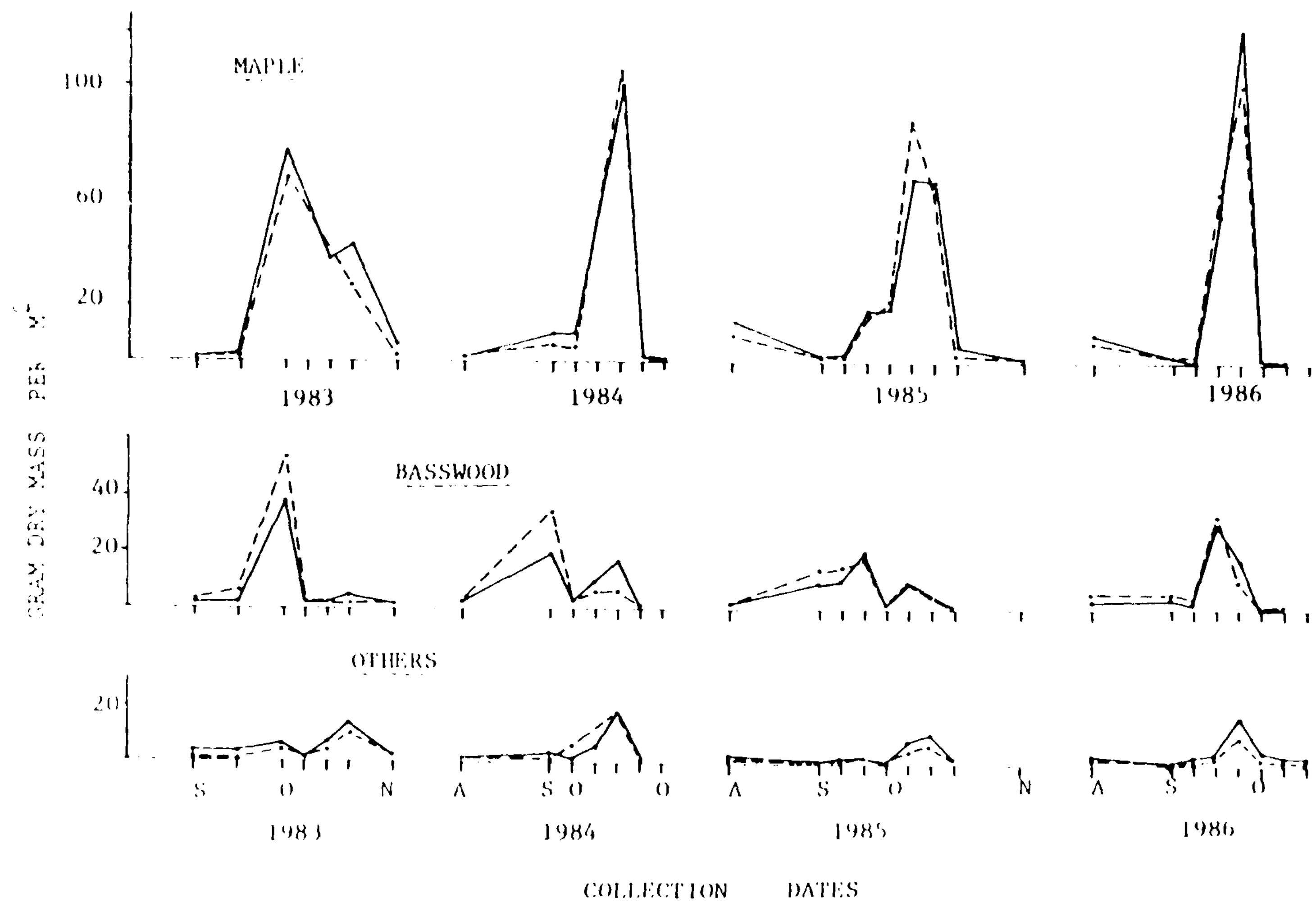


Fig. 55. Litter inputs, 1983 to 1986, in gram dry mass / m^2 (Test: broken line; Control: solid line).

3. Litter standing crops

i. Methods

As in previous years, standing crop estimates were obtained every two weeks from two sources:

a) litter moisture samples, 1/16 m^2, quickly cleaned of woody debris and soil aggregates in the field, weighed, oven-dried and re-weighed;

b) litter arthropod samples, 1/16 m^2, Tullgren-extracted, cleaned of non-leafy material when dry, and weighed.

Both types of samples tend to overestimate standing crops. They are contaminated with soil and animal faeces, which are impossible to detect and remove completely, especially after they have been "baked on" during drying. In 1986, we began to implement a washing method for these samples. It consists of submerging each sample inside a large screen placed into a tray filled with water, just long enough to allow the litter to become flexible. The leaves are then rubbed to clean them of soil and sand, and are fished off the surface, squeezed and bagged. The screens are turned over and hosed off into the tray. Small particles are recovered by pouring the water (soil tends to remain on the bottom) through a fine-mesh screen from which they can be scraped off, to be added to the coarse litter sample obtained in the first step.

In an effort to quantify the difference between past and present methods, both were employed on a total of 300 samples/site. Unfortunately, our facilities necessitate that samples be washed outdoors, and the onset of cold weather prevented us from processing all samples of the 1986 season. Not until May of the current year will we be able to wash the remainder, which are stored, dry, in the Upper Peninsula.

Available data indicate that washing reduces sample mass by approximately 20 to 30%. So far, we have washed only samples of litter which had been in the

field over the winter, and loss of soluble materials is likely to be negligeable. However, for samples (still in storage) taken during the period of leaf fall, we will quantify mass loss encurred by leaching during the washing process.

For 1986, we report standing crop estimates based only on litter moisture samples (not washed). Replication, ordinarily 40/ site/ date (moisture + arthropod samples) is thus reduced to 20. Data yet to be obtained will allow us to re-calculate litter decay rates for previous years, using standing crop estimates adjusted for soil contamination.

ii. Results

Unavoidably, standing crop estimates based on 20 samples/ site in 1986 (Fig. 56) were more variable than in 1985 (Fig. 57). However, fall maxima (44[illegible] g/m^2 in Control, 414 g/m^2 in Test) were significantly different neither between sites nor between years.

In 1985, we reported 95% decay periods of 2.6 years for Test and 2.5 years for Control. Again using Olson's (1963) method, we calculated 95% disappearance as occurring in 2.7 years in Test and 2.5 years in Control, based on 1986 data. We expect that standing crop estimates obtained from washed samples (averaged over years after adjutment of earlier data) will reduce these decay rates for both sites.

4. Litterbags

We reported last year that litterbags of both 1 mm and 5 mm mesh size retarded litter breakdown: after approximately one year in the field, 95% turnover time for litter in 5 mm bags was estimated at about 5 years. The same series of litterbags, placed in the field in November 1984, were again

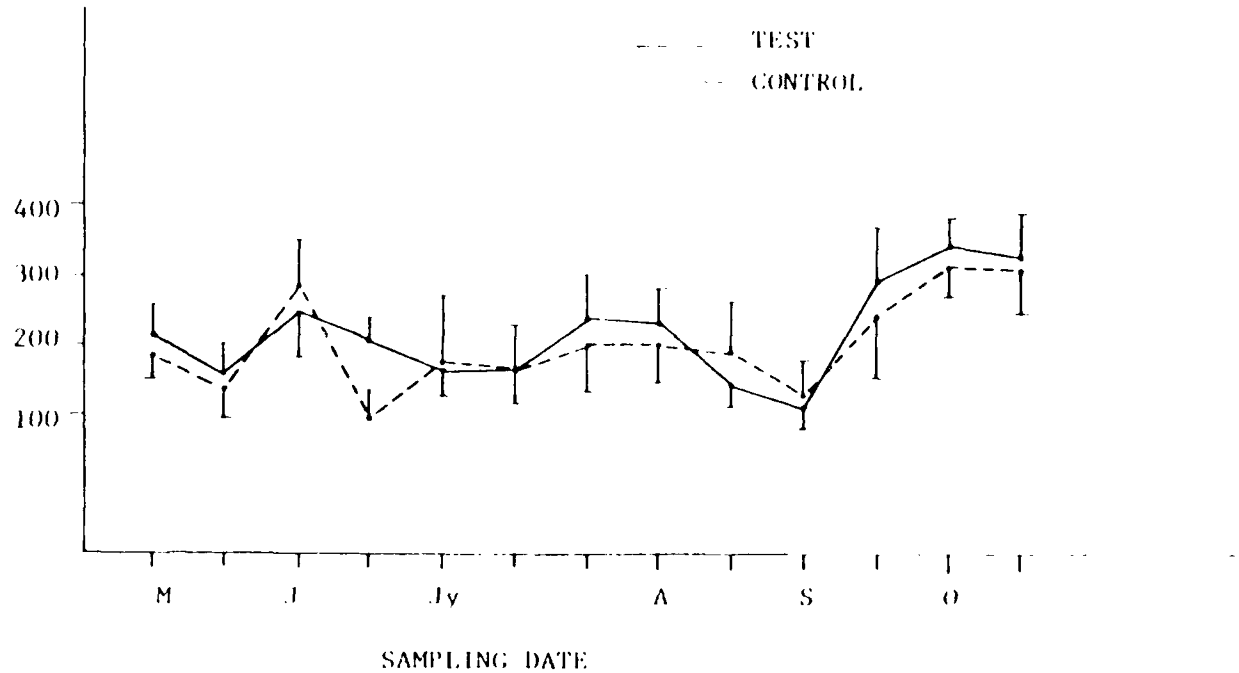


Fig. 56. Litter standing crop estimates for 1986 (N = 20), in gram dry / $m^2 \pm 95\%$ confidence limits.

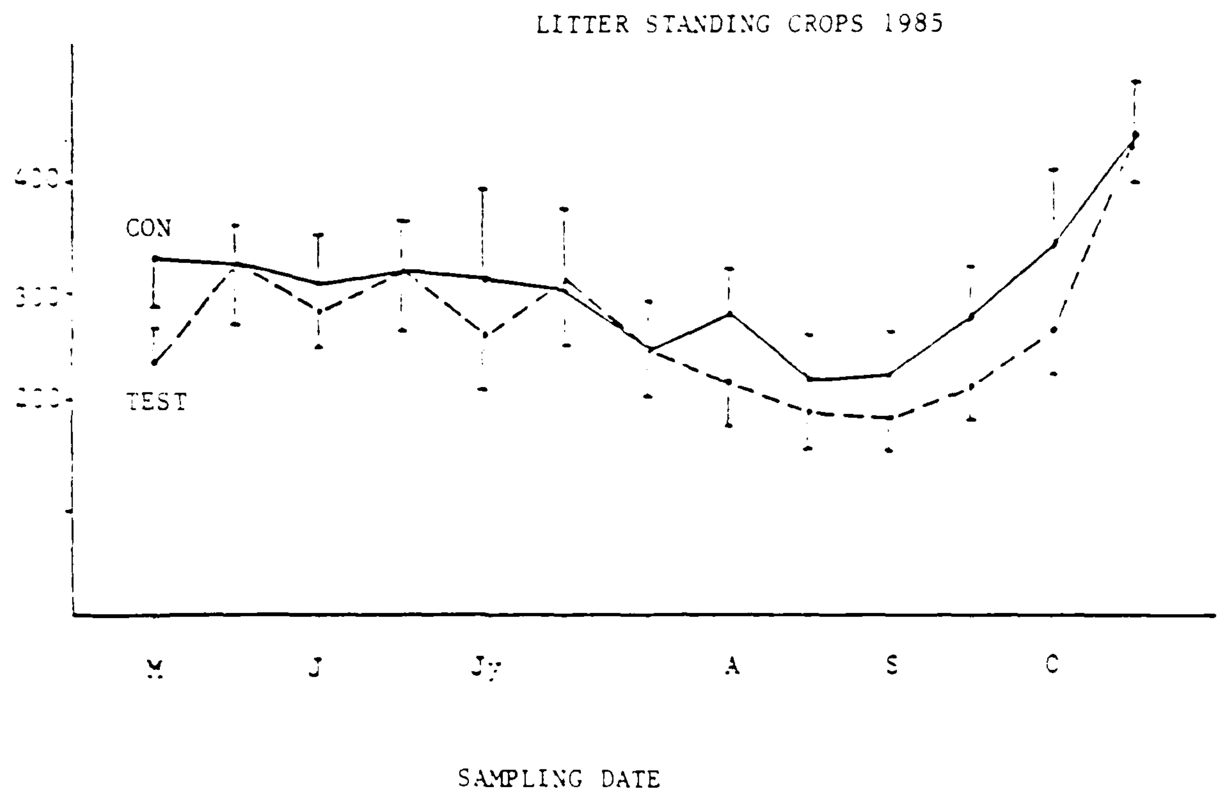


Fig. 57. Seasonal litter standing crops in Test and Control, 1985, (means ± 95% confidence limit, N = 40).

sampled in 1986, ending with a last sample on September 22, 1986. After 22 months in the field, 50% of initial mass was still retained in 1mm mesh bags, and 26 and 33% (Test and Control respectively) remained in 5 mm mesh bags. Final mass losses were identical in Test and Control for 1 mm mesh bags, and did not differ significantly for 5 mm bags.

In our sites, turnover rates obtained by the litterbag method would be unrealistic. Furthermore, obvious contamination of these samples with soil and worm casts (of D. octaedra) increasingly biases the data as time progresses. Therefore, we propose that these studies not be repeated in the future, even though they seem to provide a means of comparing decomposition rates in Test and Control.

5. Leafpacks

i. Methods

Methods for assembling, sampling and processing leafpacks have been described in detail in our last report, and have remained essentially unchanged. Briefly, six dried, pressed maple leaves are assembled per pack, placed directly on the soil surface, and secured by a loose hood of 2.5 cm mesh netting which is pinned down. After retrieval from the field, leaves are separated from each other and cleaned gently in water, oven-dried and weighed.

Series I leafpacks (3 subsets: sun, shade and randomly mixed leaves) was placed in the field in November 1984. A replicate series, II, (2 subsets: sun and shade leaves) was initiated in November of 1985.

In addition, a number of "litterpacks", consisting of air dried, loose leaves which were not pressed, confined in 2.5 cm netting, were set out in November of 1985. At the time, we had thought to use litterpacks for nutrient analyses, but have since been unable to continue this particular work element. We did,

however, obtain mass loss data at monthly intervals in 1986: approximately 52% of initial mass remained in both Test and Control after 11 months exposure in the field (Fig. 58). Again, contamination with soil and worm casts provided a source of bias. We propose to validate mass losses by washing a number of these samples in the spring of 1987. They have been stored, dry, for that purpose.

ii. Leafpack mass loss

We have so far reported only the mass loss data for one site (Control), obtained from leafpack series I started in November of 1984.

For entire packs, complete results are presented in Figs. 59 and 60. After the second winter in the field, some packs were difficult to retrieve or had apparently been trampled. What we could retrieve in sufficient replication from both sites were mixed packs (N = 14/site) in May of 1986. Following the second winter, approximately 40% remained of initial mass (Fig. 59). Means for Test and Control differed at $0.01 < P < 0.05$. Circumstantial evidence indicated that A. longa may have contributed to this difference by pulling leaves into its burrows: only in Test had entire leaves disappeared from 10 of the 14 packs, mainly those in top and bottom positions. Over the course of the first year of decomposition of mixed packs, temporary differences between sites existed, but mass loss endpoints were virtually identical (Fig. 59).

Sun and shade leaves of series I were sampled four times during 1985 (Fig. 60). Like that of randomly mixed leaves, remaining mass after approximately one year in the field did not differ between sites.

Series II leafpacks, set out November of 1985, consisted of sun and shade subsets. We increased sampling replication from 8 to 1[illegible] packs/date, and were able to obtain a late November sample (one full year in the field) in spite of

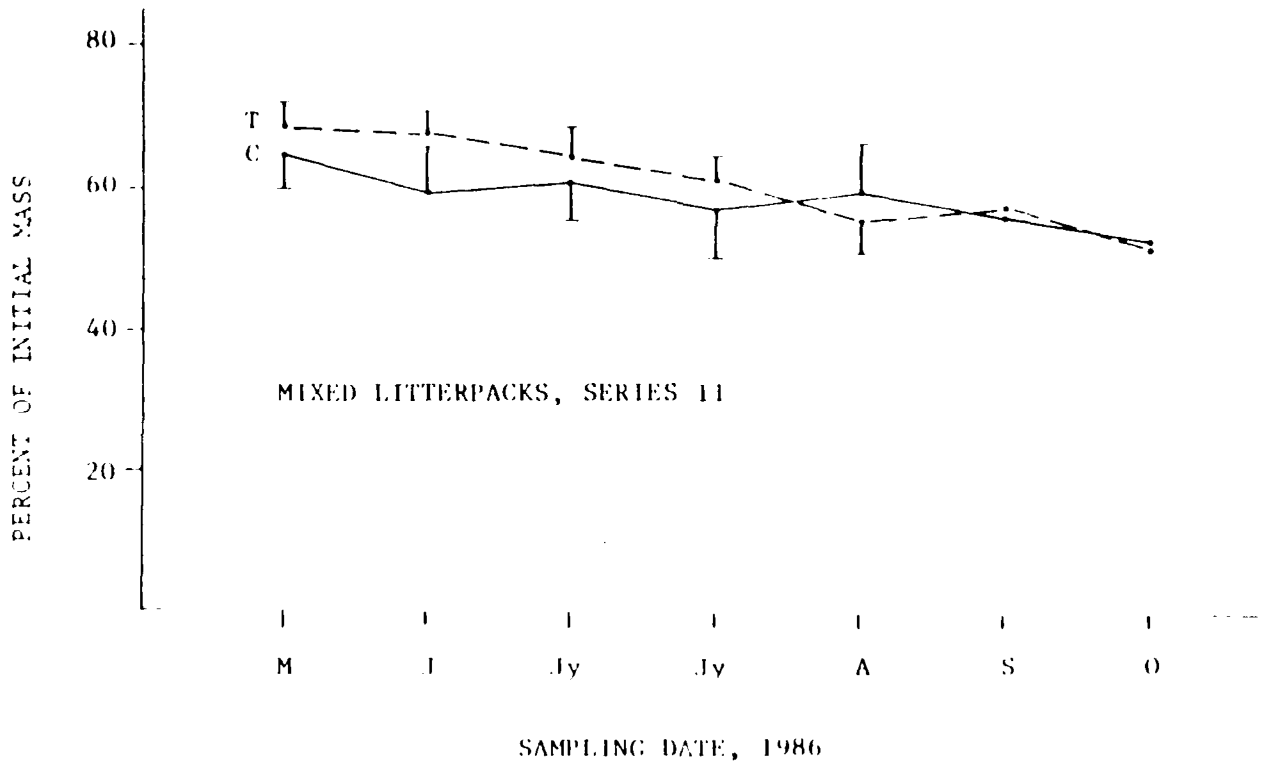


Fig. 58. Percent remaining of initial mass, for mixed litterpacks set out in November of 1985 and sampled through 1986. (Means ± 95% confidence limits).

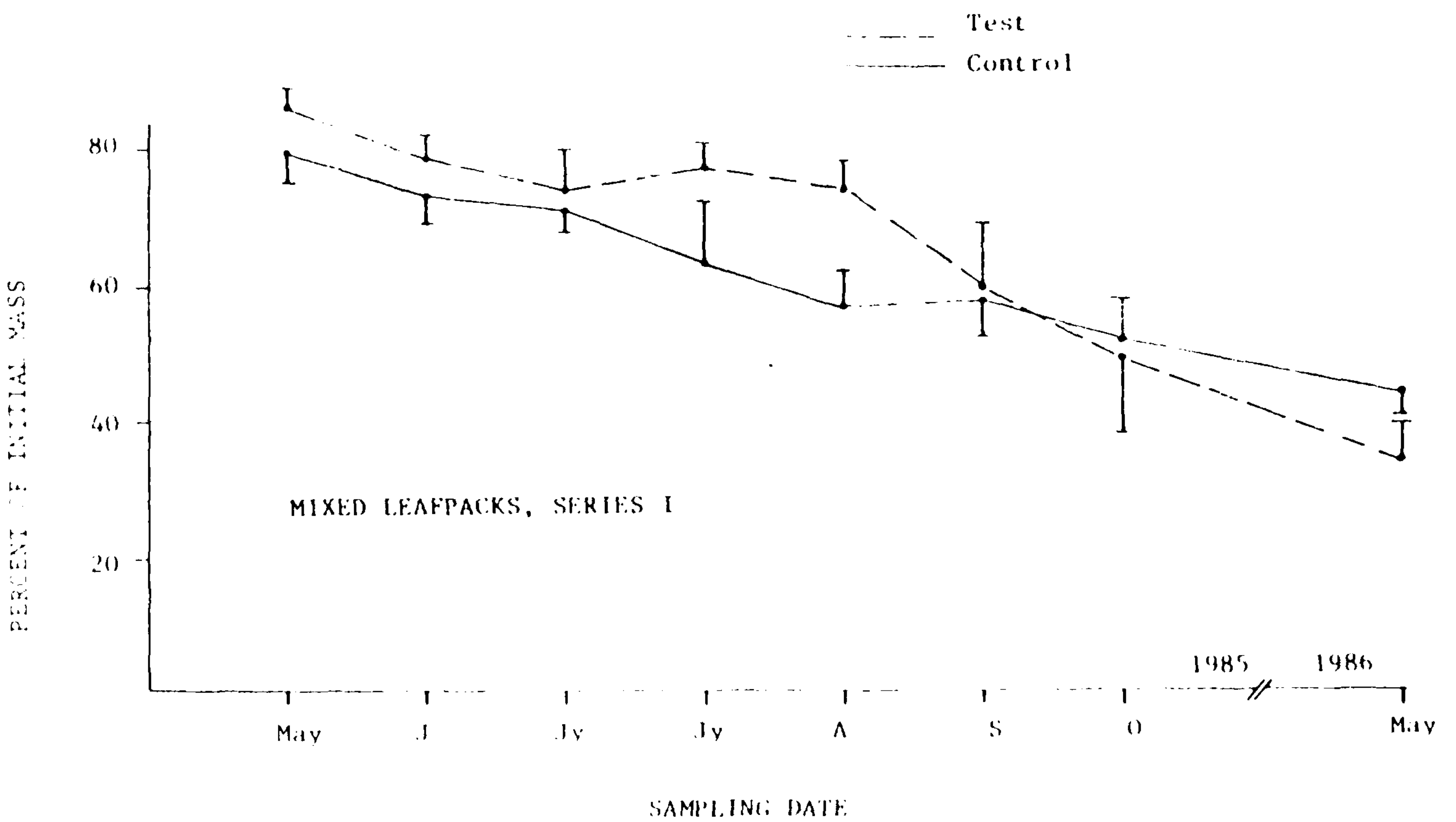


Fig. 59 . Percent remaining of initial mass, mixed leafpacks, set out November 1984; means ± 95% CL.

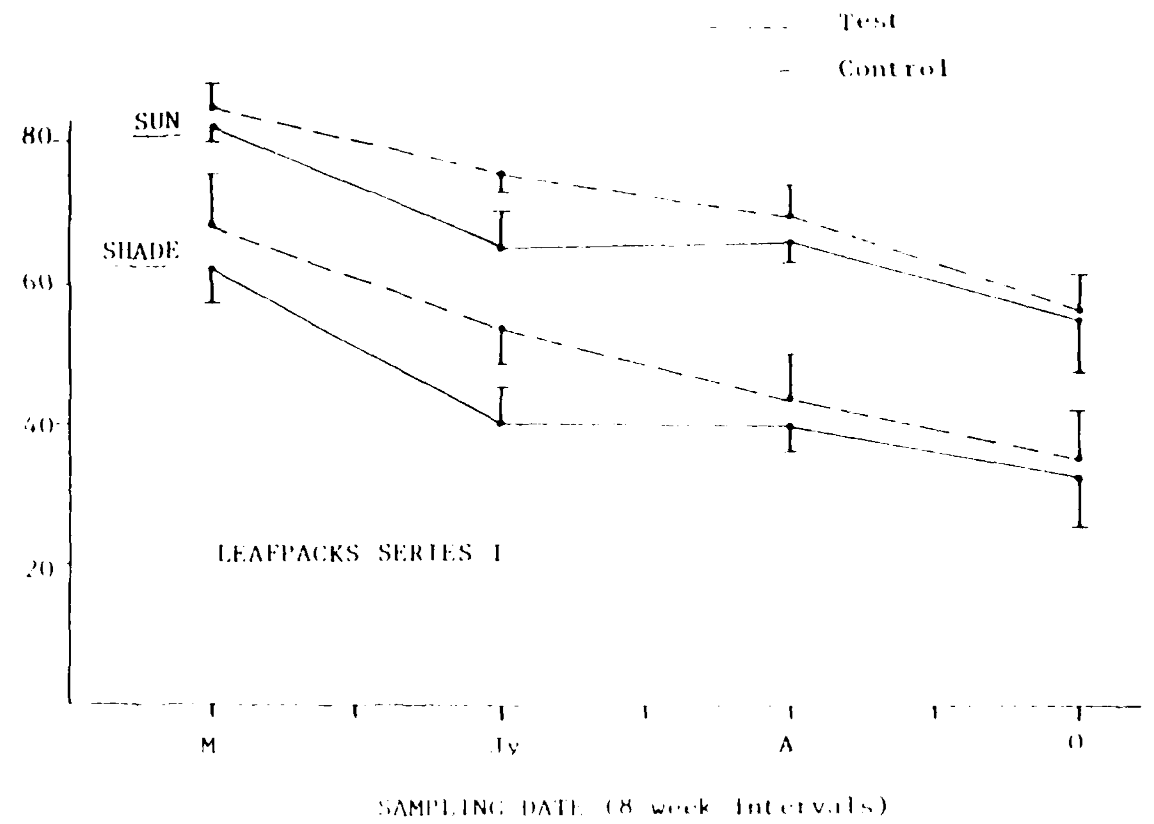


Fig. 60. Percent remaining (means ± 95% CI) of sun and shade leafpacks in Test and Control, series I (set out November 1984).

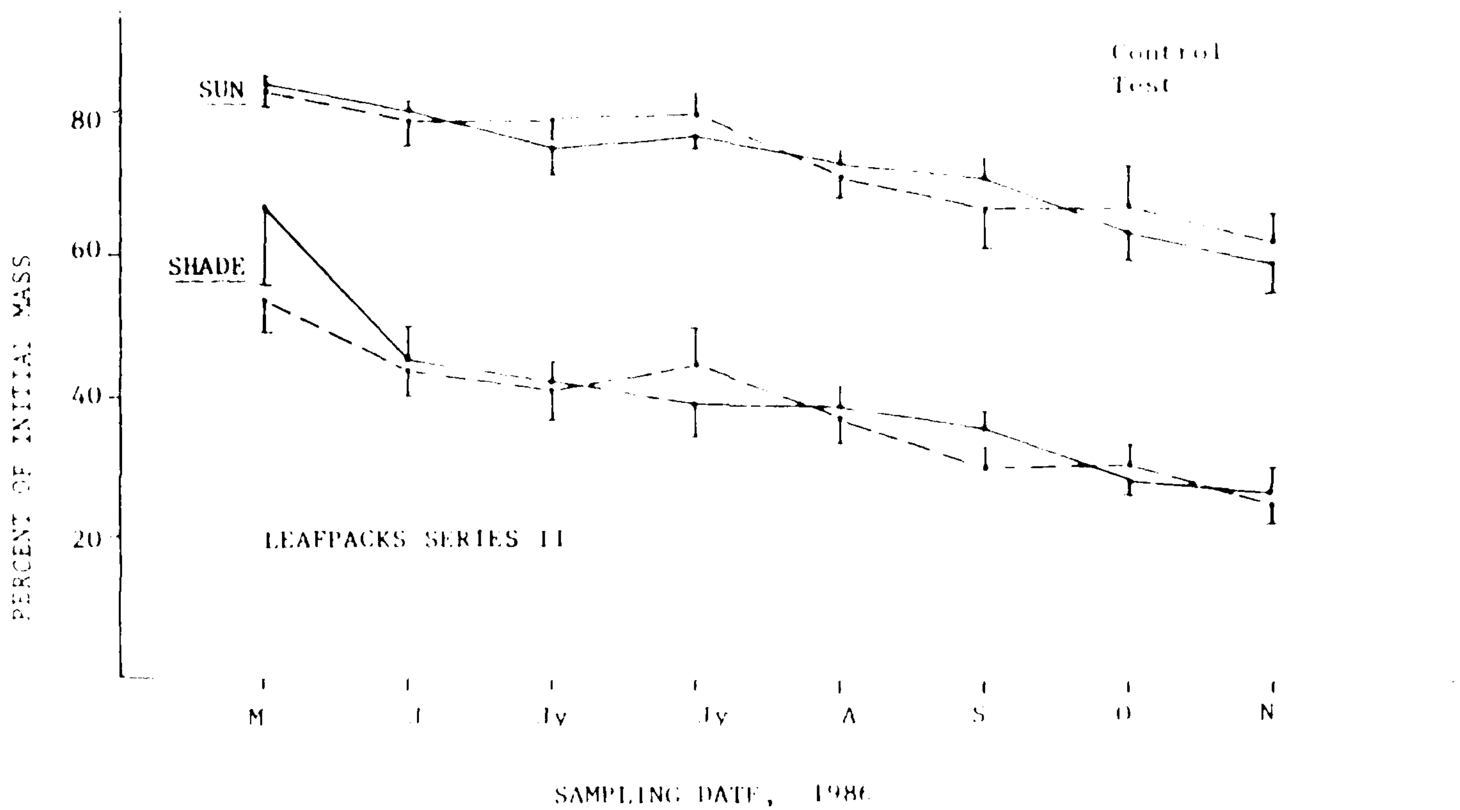


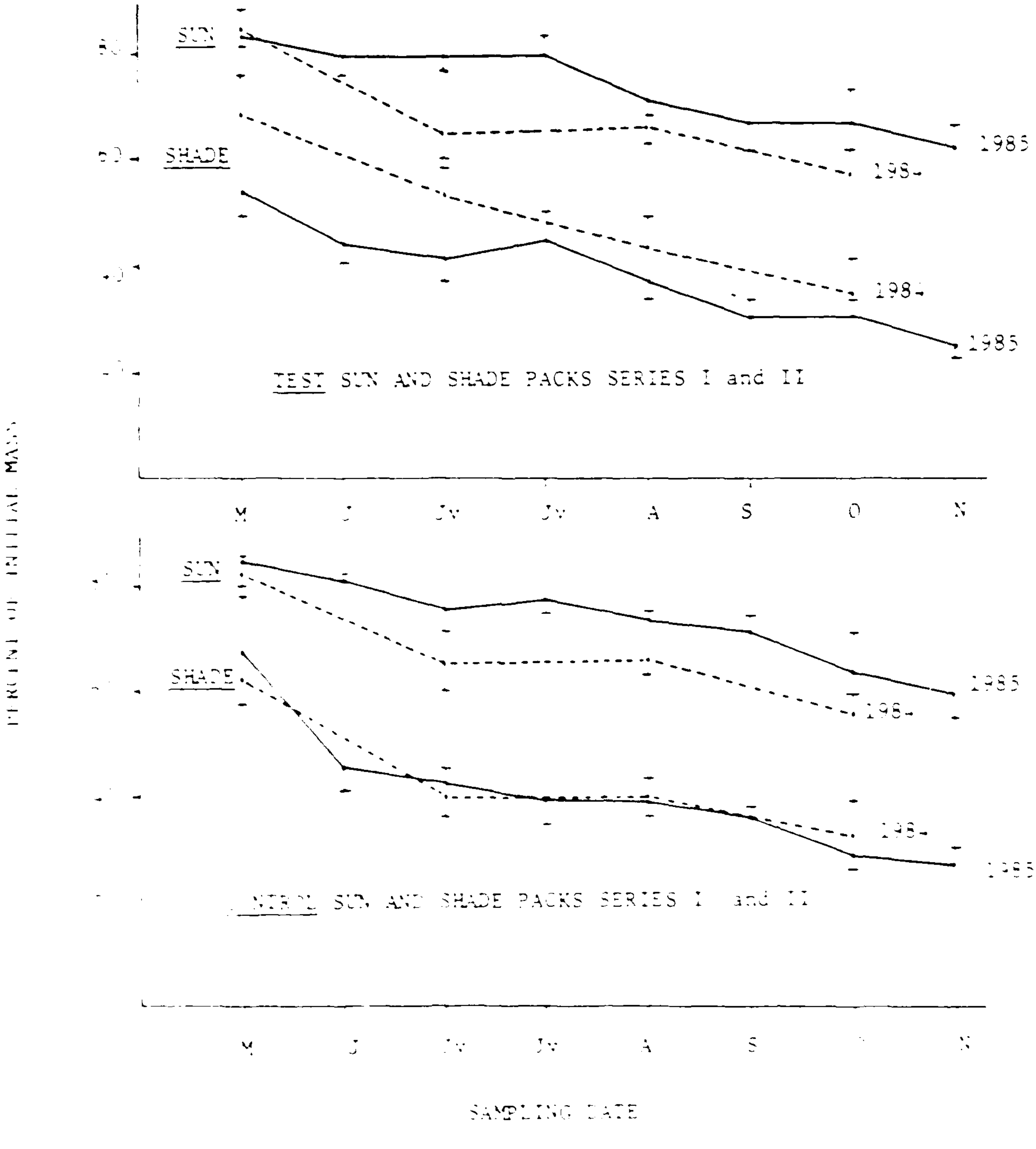
Fig. 61. Percent remaining (means ± 95% CL) of sun and shade leafpacks, series II (placed in the field November 1985).

snow cover. Slightly less than 30% of initial shade leaf mass remained at that time, while sun leaves retained approximately 60% of initial mass. Clearly, decomposition rates in Test and Control were equal (Fig. 61).

Comparison of series I and II sun and shade leaves shows some between-year differences as well as between-site similarities (Fig. 62). Sun leaves in both sites lost more mass during the summer of 1984, although October samples of series I and II did not differ significantly. Shade leaf decomposition was essentially identical in 1984 and 1985 in Control, while series I and II differed in Test during the first half of the seasons.

We have also reported differences in mass loss depending on position of a given leaf within the pack; top and bottom leaves experienced slightly greater decay than intermediate leaves (series I, Control, 1985 annual report). In series II leafpacks, these relation-ships were more variable, although the same general trends emerged. In Fig. 63, we illustrate changes in mass observed for top and bottom leaves only. In both sites, the uppermost leaves of shade packs, and the lowest leaves of sun packs, decayed to a greater extent than their respective counterparts. Although the divergence seemed great by November of 1986 (e.g., Test sun, Fig. 63),in none of the cases was it statistically significant.

In conclusion, we note the close agreement of mass loss rates between sites, particularly within the same year (e.g., Fig. 64). Any differences are partly attributable to "leaf type" of individual leaves. Leaf type is easily characterized by the ratio of surface area : mass, which is unique to each leaf. This ratio, together with the position of a given leaf within a pack, provide the main parameters for detailed analysis of breakdown rates in Test and Control.



Percent remaining (means ± 95% C.L.) of sun and shade packs of series I [illegible] and series II [illegible].

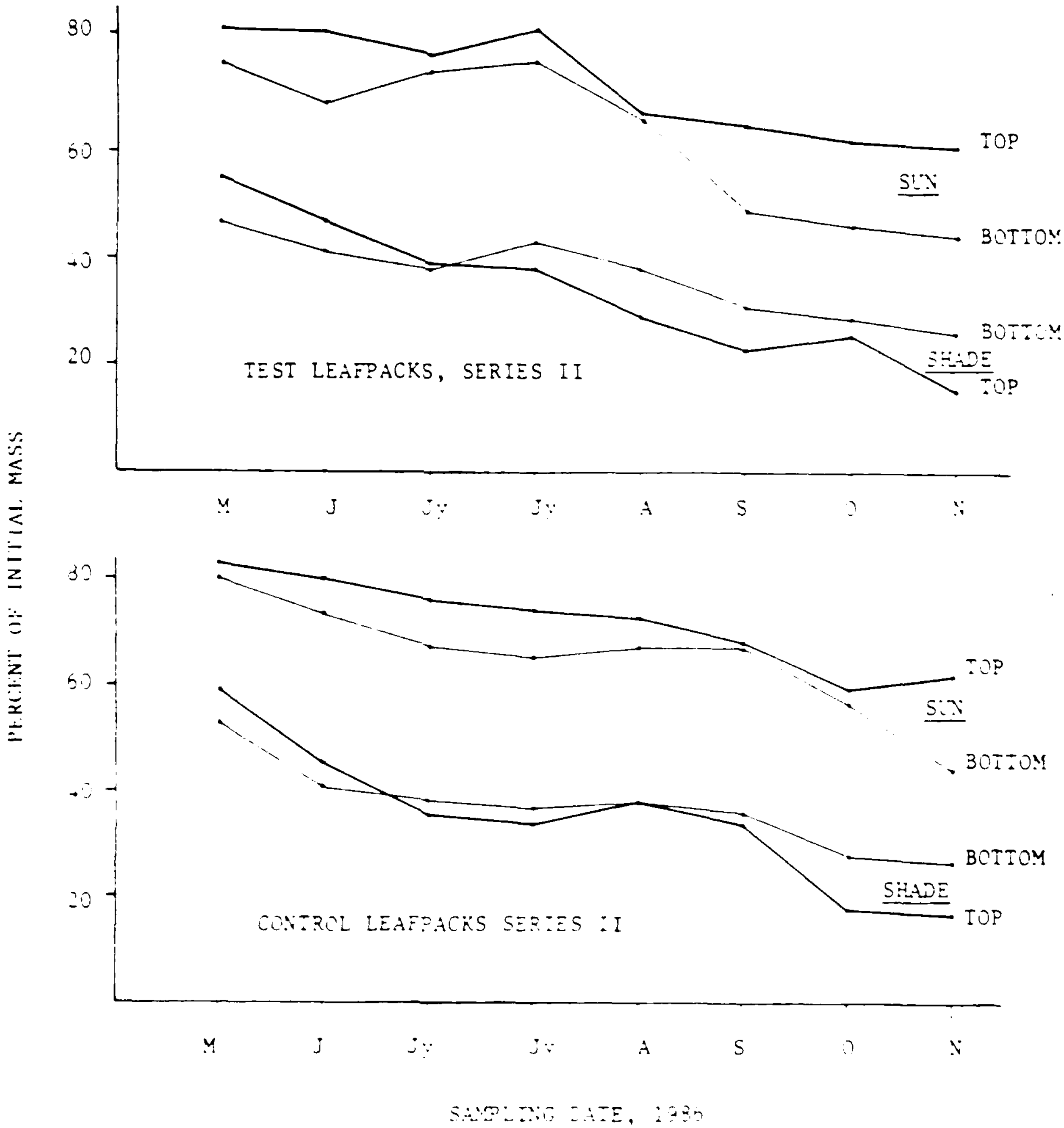


Fig. 4. Percent remaining of sun and shade leaves in top and bottom positions in Test and Control leafpacks series II (set out November 1985).

b. Statistical treatment

We are currently assembling a leafpack data file for ANOVA with sites, dates and position within the pack as main factors, and initial area: mass ratio as the single covariate. Results will answer the following main question: Given a certain initial area: mass ratio, will leaves in a given position experience equal mass loss over time in Test and Control?

Depending on these results, the model may be expanded to include years as additional main factors, which would make it suitable for eventual testing of ELF effects.

For litter input data, we will continue to use ANOVA using sites and years as main effects, based on total yearly litter input per litter trap (N = 20 traps site).

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ELF Communications System Ecological Monitoring Program

BIOLOGICAL STUDIES ON POLLINATING INSECTS: MEGACHILID BEES

Annual Report 1986

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

I ABSTRACT

High voltage transmission lines and magnetic fields have been shown to affect honeybee reproduction, survival, orientation, and nest structure. ELF EM fields could have similar effects on native megachilid bees.

Two species in the genus *Megachile* have been most abundant in artificial nests at experimental and control sites in Dickinson and Iron Counties Data on their nest architecture, nest activity, and emergence/mortality have been collected since 1983. Five hypotheses concerning the possible effects of ELF EM fields are considered using these data. Although some data are not yet analyzed, what has been analyzed shows no indication of differences between experimental and control sites prior to operation of the ELF antenna. A number of changes in protocol are proposed for future research to facilitate detection of any differences once the antenna becomes operational. Sample sizes of less than 30 bees per site should be sufficient to detect reasonable differences between experimental and control areas in future studies. Such sample sizes should not be difficult to obtain.

II PROJECT RATIONALE AND OVERALL OBJECTIVES.

Effects of high voltage transmission lines and fluctuations in the earth's magnetic field have been reported to affect honeybees (Greenberg et al. 1981; Gould 1980). In addition, honeybees have been shown to have an organ in the abdomen that could be used to detect the earth's magnetic field and thus could be used as a compass in orientation (Gould et al. 1978) Because such effects of electric and magnetic fields have been demonstrated, it is possible that ELF EM fields may alter a bee's ability to orient or may otherwise affect its behavior.

Honeybees, however, are rare in the state forest where the Michigan ELF antenna is located, and are unable to overwinter in the harsh climate of Michigan's Upper Peninsula (Fischer, 1983 Annual Report). Therefore, native bees are a better choice for ecological studies of the resident bee fauna. Native bees are particularly important in ecological communities such as those in the vicinity of the ELF antenna because they are pollinators of flowering plants, and are therefore important to the reproductive success of these plants.

With the exception of bumblebees and some halictids, native bees are solitary, meaning that each female constructs and provisions her own nest rather than having a special queen caste responsible for reproduction. Solitary bees have several advantages for ecological studies. As "mass provisioners", they create a discrete cell for each offspring, and fill it with a provision mass of pollen and nectar prior to laying the egg. The bee does not add more provisions after the egg is laid. A series of such cells, each with a provision mass and egg, are created in succession by each female. The provisions that go into each cell are a direct measure of parental investment in an offspring (Strickler, 1979). The size of the adult bee that emerges from each cell is correlated with the amount of provisions provided it, and with the size of the cell in which the larva develops (Krombein 1967; Klostermeyer et al. 1973; Torchio and Tepedino 1980). However, there is a tradeoff between the investment per offspring and the rate at which offspring are produced. The more the bee invests per offspring (ie, the larger the offspring), the fewer offspring she will produce. If bees are disoriented, agitated, or slower at foraging, they may invest less per offspring, produce fewer offspring per unit time, or both. Solitary bees are unusual in having this direct relationship between parental investment per offspring, adult size, and reproductive output.

The nesting biology of some species of solitary bees in

the family Megachilidae is especially easy to study because they accept artificial nests in the field. These bees typically nest in abandoned beetle bores in dead logs. "Trap nests" of drilled blocks of wood are also used by bees as nest sites. Such artificial nests can be placed in habitats where bees are expected to nest, in order to increase the sample of nests available for study, and to standardize such characteristics of the nest as bore depth and diameter (Krombein, 1967). Trap nests are used in the management of the leafcutter bee, Megachile rotundata, for pollination of alfalfa (Hobbs, 1972). Thus there is an extensive (though unreviewed) literature on megachilid biology.

Research on the effects of high tension wires and magnetic fields on honeybees suggests working hypotheses on which to base our initial analyses of native bee nesting biology. Of possible relevance to megachilid behavior are an alleged greater tendency for dispersal, and greater levels of activity (Wellenstein, 1973), as well as reduced reproductive output, lower overwintering survival, and modifications of nest structure (Greenberg et al., 1981) when colonies were exposed to electromagnetic fields from high voltage transmission lines. In addition, disorientation due to fluctuations in ELF magnetic fields is possible if megachilids share the honeybee's ability to detect magnetic fields. (Gould et al., 1978, 1980; Gould 1980; Tomlinson et al. 1981).

III METHODS AND TYPES OF DATA COLLECTED

The hypotheses discussed later in this report are necessarily constrained by the types of data that have been collected for the past four years. These include nest architecture and nest orientation, emergence/ mortality data, and nest activity. The first two types of data are obtained by placing trap nests in the environment, and allowing bees to construct nests in their choice of traps during the summer. The following spring, various parameters of their nest architecture are measured. Bee and parasite emergence and larval and pupal mortality are recorded at the same time. Nest activity data are gathered during the summer season while the bees are constructing their nests.

Trap Nesting Methodology

Trap nests consisted of elongate white pine pieces 19x19x153 mm. drilled lengthwise to a depth of either 142mm (smaller diameters) or 107mm (larger diameters). Six different bore diameters were used. "Blocks" of 12 nests, two of each bore diameter, were bound together so that half of the nest entrances faced one direction and the other half faced the opposite direction (Fig. 1). The bore diameters were arranged randomly in the blocks, with these directional constraints.

"Hutches" consisting of a frame with four shelves and a roof were used to hold the blocks of trap nests (Fig. 2). Four blocks of nests were placed randomly on each shelf, making a total of 192 nests present at any one time. The hutch was open on both sides, so half of the nests opened in each direction.

Four study sites were selected in 1984 for placement of hutches. Two are experimental sites along the ELF antenna: Ford 1 and Ford 2 (F1 and F2), and two are control sites: Camp 5 and County Line (C5 and CL). Further information on the study sites can be found in the 1985 annual report. Three sets of two hutches, making a total of six hutches were placed at each of the four study sites. In each set of two hutches, one hutch was oriented in a north-south direction so that its nests open to the east or west, and one hutch was oriented in an east-west direction so that its nests open to the north or south. The two hutches in each set were placed close together in edge habitats between open areas where there are abundant flowering plants, and woods where natural nest constructing materials are available.

When a nest was occupied by a megachilid bee, it was

given a number that included site, hutch direction, bore direction and shelf height. Position on the shelf and in the block of nests was not recorded. When the nest was completed, it was removed from the block, and replaced with a nest of the same bore size. Completed nests were brought to Channing to overwinter, in order to avoid vandalism and marauding animals. Each nest was stored in a large test tube with cloth covering the opening. Tubes were placed in wooden boxes built to fit the hutch shelves. The following spring tubes were checked daily for bees that had emerged from the nest and were in the tubes. Date of emergence, species, and sex of offspring were recorded. Finally, adult bees were released at the sites where their nest had been constructed the previous summer. Nest architecture measurements were sometimes made before the bees emerged and sometimes after emergence, depending on year and species.

Bee species and sample sizes.

The five most abundant species that were found at the study sites between 1983 and 1986 are *Osmia tersula* Cockerell, *Hoplitis albifrons* (Kirby), *Megachile pugnata* Say, *M. inermis* Provander, and *M. relativa* Cresson. Less abundant representatives of the genera *Osmia* and *Hoplitis* also constructed occasional nests. Tables 1-4 indicate the number of nests made by the five most abundant species each year. Numbers of 1986 nests for each species have been estimated, but these must overwinter before they can be opened for nest architecture measurements and species confirmation. Also indicated in the tables are the number of hutches for which at least 5 nests of a given species were constructed. This number should be compared with the total number of hutches for the site; 6 in 1985 and 1986, fewer in 1983 and 1984 (see top of the table) because sites were being selected and protocol was being developed. Hypotheses about the acceptability of different nest orientations can be tested most easily for species with at least 5 nests in all hutches.

M. relativa is the only species that consistently constructed large numbers of nests at all sites, and that usually had over 5 nests per hutch. *Osmia, Hoplitis, M. pugnata*, and *M. inermis* constructed few nests at some sites, and less than 5 nests per hutch at many hutches. Furthermore, the *Osmia* and *Hoplitis* tend to have nests with few cells, and are highly variable in the presence and size of unoccupied interstitial, basal and vestibular spaces. This variability will make changes due to ELF EM fields hard to detect. Furthermore, identification of the species that constructed *Osmia* and *Hoplitis* nests is difficult for the untrained eye, especially if no adult bees emerge from the nest in the spring.

We conclude that M. relativa is most suitable for our analyses of nest architecture. However, we are not yet prepared to eliminate M. inermis from our study, because most of the nest activity data collected to date are for this species. M. inermis has a number of advantages for behavioral observations. Their large size and relatively slow movements make them easy to observe both at the nest and in the field. It is easy to distinguish whether they have returned to the nest with round leaves (for cell caps), oblong leaves (for cells), or pollen (see "Nesting biology of a megachilid bee", below). Furthermore, they are the only species that continues to forage during rain showers (V. Scott, personal communication), when the sun is obscured by clouds. Sun compass orientation is difficult at this time so magnetic field orientation may be important.

In our future research we propose to attempt to increase the nesting populations of M. inermis at the control sites. By focusing on only two species and thus using fewer bore sizes, we will increase appropriate bore sizes for M. inermis at the hutches. However, we doubt that appropriate bore sizes were limiting at the hutches in the past. Observations in previous years suggest that M. inermis accepts Circium flowers more readily than other plant species. Members of this genus are rare at the CL site. Transplanting Circium flowers may increase the M. inermis population. Additional bees may be introduced to the control sites from other areas in hopes of increasing the nesting populations at the hutches. As a last resort we may consider alternat[illegible] control sites from among those that are already being [illegible] other ELF projects.

In conclusion, our study in the future wi[illegible] the two Megachile species, M. relativa and M[illegible] analysis of the data for these two spe[illegible] complete. This report is primarily a summa[illegible] of our analysis.

Nesting Biology of Megachilid Bees

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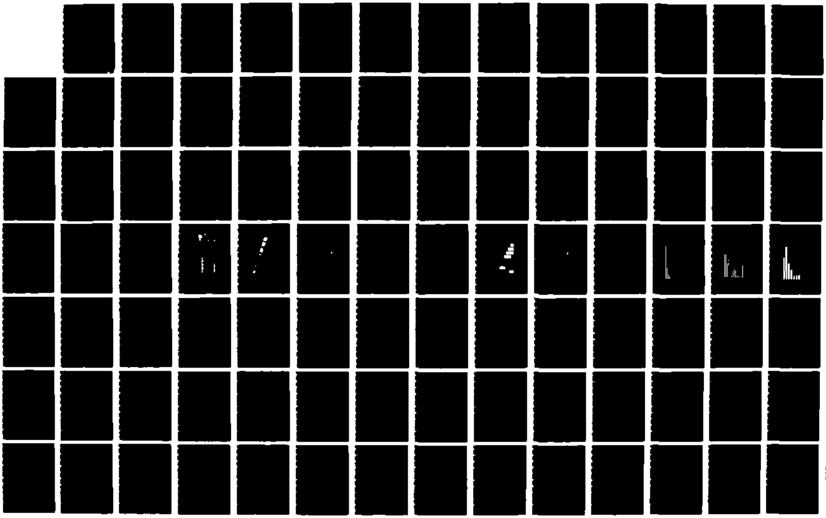

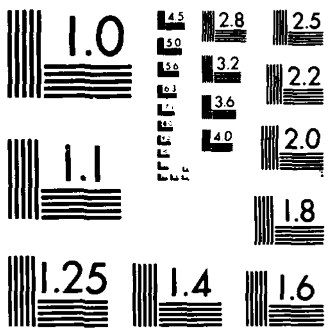

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base of the first cell. Next she cuts and brings to the nest several elongate pieces of leaf in succession. These are used to line a tube- or cup-shaped cell that is slightly longer than her body. Next she makes a series of pollen and nectar foraging trips to fill the cell with the discrete provision mass that will be the larva's food supply. When provisioning is complete, the female lays an egg. Fertilized eggs become a females while unfertilized eggs become males. The female has voluntary control over the sex of the egg that she lays (Klostermeyer and Gerber, 1970). After laying the egg, she cuts more round leaves to cap the cell, sometimes adding chewed leaves, sand, pebbles, or bits of wood to separate the cells. Next she cuts more elongate leaves for the second cell, and repeats the process. Thus a linear series of cells is constructed in the nest bore. Typically, the cells at the base of the nest are more likely to contain females and the cells near the entrance are more likely to contain males (Krombein, 1967). Since females are typically larger than males in these bees, cells at the base of the nest tend to be larger than cells at the entrance. When she has completed the last cell that she is going to put in the nest, she constructs a series of plugs of round leaves, chewed leaves, and possibly other material. Sometimes there are empty "vestibular" spaces between segments of plug. Sometimes there is one long mass of plug material. There may also be space between the outermost plug and the opening of the nest, called an "indentation".

Each female may construct several such nests over her life time. Some nests are abandoned before they are finished because the bee has died, or for other unknown reasons.

Inside each cell the egg hatches, and the young larva feeds on the provisions prepared by its mother. Both Megachile species in our study are univoltine in Northern Michigan, and both overwinter as prepupae. Pupation occurs in the Spring, and the adults emerge soon after, in mid-June at our study sites. A variety of parasites may emerge from the cell instead of the original bee. Oviposition of parasite eggs usually occurs while the cell is being provisioned, when the mother bee is out of the nest on a pollen foraging trip.

IV NEST ARCHITECTURE DATA

Condition of past data

1983 nest architecture for M. relativa is available on data sheets and was entered into the computer before the recovery team began work. The computer files are being edited so that they are in the same format as the 1985 data (see below), as far as is possible. The original 1983 nest architecture data sheets for M. inermis have been lost.

Some 1984 nest architecture for the Megachile species is on data sheets, but much is incomplete and unverified. The reliability of cell sizes and species identifications are considered to be low for many of these nests because of the inexperience of the individual who filled out the original data sheets. No measurements were taken on many of the nests, and no emergence data were taken on others. The original nests from which architecture data were measured have been lost or discarded. If information is retrievable from the 1984 data on Megachile nest architecture, it will be analyzed at a later date.

Although emergence from 1985 nests was recorded in early summer of 1986, nest architecture measurements were not made until November and December, 1986. These data were added to the computer data base in January, and were analyzed in February and March. 1985 nests for M. inermis have not yet been measured.

Nests constructed in 1986 will not be opened until the spring of 1987, so no data are yet available about their nest architecture.

Measurement

Three people measured the 1985 M. relativa cells. Each person measured every third nest so that any biases in measurement would be distributed evenly between sites and dates.

Nests were split open lengthwise with a chisel. Non-reproductive spaces (basal space, vestibular spaces, associated caps, nest plugs, and indentation) were measured with the cells intact. Each cell was then removed and measured from the base of the cell to the position of the outermost leaf in the cell cap (Fig. 4). Since nest architecture measurements for 1985 nests were taken after emergence, most cell caps had been destroyed and were not measurable, although a ring of leaf indicated where the cell

cap had ended, even if the cap itself was not present. If the occupant died before emergence, then the cell space and cell cap were measured separately (Fig. 4).

Cell volumes were calculated using cell length and bore diameter measurements, and assuming that the cells were cylindrical.

Hypotheses involving nest architecture data.

Hypothesis 1: The average size (length and volume) of cells for each offspring, and/or the average number of cells produced per nest will be altered by exposure to ELF electromagnetic fields.

Rationale

Honeybee reproductive output decreased on exposure to high voltage transmission lines (Greenberg, et al., 1981). ELF fields may have a similar effect on megachilids. The ELF electromagnetic fields may affect both cell size and nest architecture in various ways. For example, if bees are disoriented by the fields, they may gather resources (leaves, pollen) more slowly when exposed to the fields than when not exposed. As a result, they may produce new cells at a slower rate, or they may produce smaller cells.

Previous studies have found that the weight of offspring of the generalist megachilid, Osmia tersula, is lower if their cells were produced late in the season rather than early in the season (Torchio and Tepedino, 1980). This species also showed an increase in the proportion of male offspring (the smaller sex) produced late in the season. A reduction in offspring size late in the season is thought to be related to reduced foraging rates due to aging of the bee (Torchio and Tepedino, 1980, Tepedino and Torchio, 1982). Similarly, ELF EM fields may slow the foraging of M. relativa and M. inermis, resulting in smaller bees produced in smaller cells. A size reduction could affect cells with offspring of both sexes, or it could reflect the production of a greater proportion of male offspring, for species with smaller males than females.

In contrast to the generalist megachilids, the pollen specialist Hoplitis anthocopoides did not show a reduction in offspring weight late in the season, in spite of reduced foraging rates (Strickler, 1982). Rather, it was hypothesized that slower foraging rates led to fewer offspring per nest late in the season as compared with early in the season for this species. Similarly, M. relativa and M. inermis may produce fewer cells per nest in response to slow foraging rates due to ELF EM fields.

In testing hypothesis 1 we are interested first in determining whether there are differences between experimental and control sites in cell lengths, cell volumes, and number of cells per nest. Ideally, we hope to find no differences between experimental and control sites, and between years, before the ELF antenna is operational. Then, if significant differences between experimental and control sites do appear after the antenna is turned on, we can attribute these differences to the effect of ELF EM fields. Secondarily, we will examine the contribution of a number of other factors such as nest diameter, date that the nest was begun, and the offspring's sex, to the variance in cell length and volume. Ideally, we hope to find some contribution of these factors to variability in cell lengths, cell volume, and cells per nest before the ELF antenna is operational. If factors such as aging of the bee or sex ratio contribute to the variance now, then changes in these factors due to ELF EM fields are possible. Such changes will be the underlying cause of differences between treatment and control sites in cell length, cell volume, and cells per nest. For example, if sex of offspring contributes significantly to the variance in cell lengths before the antenna is operational, then cell lengths could decrease after the antenna is operational because a higher proportion of male offspring are produced. An understanding of the contribution of a variety of factors to the variance in cell lengths, cell volume, and cells per nest will also help us decide whether changes in protocol are needed in future seasons.

Results: Cell lengths and cell volumes.

Analysis of variance was used to test for the significance of a variety of factors on cell lengths and cell volumes from all 1985 M. relativa nests. These factors were nest diameter, experimental vs. control areas ("exp"), site nested within experimental or control areas ("site[exp]"), complete vs. incomplete nests, date on which nest was begun ("date begun"), number of cells per nest, and cell order. In addition, we considered two factors that relate to our measurement techniques: person measuring (doneby), and whether or not a cap was included in the measurement of cell length (caplflg). Incomplete cells (cells for which a cap was not constructed) were eliminated from the analysis. Our first analysis treated each cell as an independent measurement. This allowed us to test whether cell order contributes significantly to the variance in cell lengths and volumes as would be expected if females are more often found in basal cells than are males. We used the GLM procedure of SAS to calculate Type IV mean squares; all variables used overall model error as the error term except exp, which used

site[exp] mean square as an error term.

All factors contributed significantly to the variance in cell length except experimental vs. control sites, and complete vs. incomplete nests (Table 5). The same was true for cell volume, except that number of cells per nest was also not significant. Because experimental and control sites were not significantly different, we are encouraged that differences between the two types of sites after the antenna is operational will be attributable to ELF EM fields. We caution, however, that it will be necessary to analyze nest architecture results from 1983 and 1986 to be sure that experimental vs. control sites consistently show no significant differences in cell lengths and volumes.

The effect of bore diameter

The overall variability in both cell length and volume is low (CV < 10% of the mean). More of the variance in cell volumes than in cell lengths was explained by the factors tested (r^2 = .89 and .23 respectively). This difference is primarily due to the greater correlation of nest diameter with cell volume than with cell length, as illustrated for basal cells (C1) in Figures 5 and 6.

In 1985 M. relativa constructed nests in bores made with four different drill bit sizes. Nest diameters varied considerably within a bore size, so that bore sizes overlap in actual diameters. Though nests of a given bore size were all drilled with a bit of the same diameter, the actual bore diameters can vary somewhat in the drilling process, and may change due to shrinkage or expansion of the wood when exposed to the elements. Figure 6 indicates the distribution of bore diameters used by M. relativa for each bore size (note that bore sizes are not ordered by mean bore diameter; size 3 is supposed to be a larger diameter than bore size 5. The few nests made in bore size 3 were in bores that overlapped in diameter with bore size 5).

The distribution of bore diameters used (all sites combined) was skewed slightly toward larger sizes (Figure 7). This is probably because a bee can always use a nest that has a larger diameter than her body, but not one that has a smaller diameter than her body. Bore diameters between 4.75 and 6.25mm appear to be most acceptable to M. relativa (Figure 7). Bore size 4, made with a bit 5.6 mm in diameter, encompasses this range. In future research we will restrict ourselves to nests of bore size 4 for attracting M. relativa. Nests of bore size 7, made with a bit 11.1 mm in diameter, (the largest size) will also be used for attracting M. inermis. Variability should be reduced by providing nests of

only one bore size for each species.

The weak negative correlation between nest diameter and cell length, and the strong positive correlation between nest diameter and cell volume suggest that cell lengths are more or less constant, whereas cell volumes increase with increasing bore diameter. We hypothesize that a bee uses her body length as a guide for cell size. In the next fiel season we may try to test this hypothesis by correlating wing length or head width (which scale with body size) of mother bees that are constructing nests with the length of the cells in the nest.

If cell length determines cell size, then why study cell volume? This depends on whether the provisions in a cell (and thus offspring size) correlate best with cell length or with cell volume. The easiest way to answer this question is to weigh the bees that emerge from each cell and correlate these weights with both length and volume of the cell from which the bee emerged. While some data on offspring weights were taken in past years for M. relativa and M. inermis, these cannot always be correlated with a specific cell. To make such a correlation will require keeping individual cells separate for emergence, so that offspring weights and cell lengths can be matched. This will be possible if nests are measured shortly before emergence, and if each cell is kept separate for emergence. In the future, instead of measuring nests after emergence as we have done with the 1985 nests, we propose returning the nests to the laboratory in late spring, about three weeks before emergence, in order to get nest measurements. Cells will be separated, leaving the leaf lining intact to minimize mortality due to handling. Each cell will be measured, weighed, and then stored separately in a vial until the bee or parasite emerges. Species and sex of the adult will be recorded before release at the appropriate field site. The leaf remains of cells from which Megachile emerged will then be reweighed. The difference in weights before and after emergence will estimate the weight of the bee that emerged, without unduly stressing the bee.

The effect of including vs. excluding cell cap length

Cells with cell caps included were slightly (but significantly) longer on average than cells with cell caps excluded. However, summing the cell and cap lengths for cells with caps excluded yielded total lengths that are slightly longer and more variable than cells measured with cell caps included. This is because there are two measurement errors in the summed length, but only one measurement error for cells measured with caps included. In the future cells will always be measured so that cap length

is included. Caps will be measured separately if present (Figure 8).

The effect of cell order

As mentioned earlier, trap nesting species usually produce cells that contain females near the base of the nest (inner cells) and cells that contain males near the entrance (outer cells). In species with males smaller than females, the outer cells tend to be smaller than the inner cells. A few known exceptions to this pattern are species with territorial males that are larger than the females. In such species, inner cells tend to contain males and outer cells tend to contain females.

Based on a few weights of offspring from previous years, M. relativa females are 1.4 fold larger than males (Table 6). Larger female cells at the base of the nest could explain the significant contribution of cell order to variance in cell lengths and volumes (Table 5). M. inermis shows more size dimorphism than does M. relativa so cell order should contribute more to variance in cell length and volume for the larger species than for the smaller.

Duncan's multiple range test was performed on mean cell length for cells in different positions within the nest (Table 7) The basal cell (C1) is significantly larger than the 10th cell (C10), but intermediate cell lengths were not significantly different. C1 had the highest proportion of female offspring of all the cells (Table 8).

Number of nests to detect differences in cell lengths and volumes.

In order to estimate the number of nests needed to detect differences between experimental and control sites in cell lengths and volumes, we calculated an ANOVA on mean cell length or volume per nest, weighted by the inverse of standard error per nest. If a nest consisted of only one cell, the inverse of the overall standard error of the model was used as a weighting factor. We restricted this analysis to nests in bore size 4 and cells with caps included in the length so that these data would be most comparable with our future protocol. Nest diameter contributed significantly to variance in mean cell lengths and volumes per nest, in spite of the reduced range of diameters tested (Table 9). No other variables made a significant contribution to variance.

Using a CV of 8.5 mm (Table 9, cell volumes), we estimate that 24 nests for both the experiment and control areas will be required to detect a 10% change in cell length

with a power of .9 and an of .01. We should be able to reach this sample size for M. relativa.

A 10% change in cell length amounts to slightly over a millimeter. This value needs to be compared with the accuracy with which cells can be measured. Mean cell length differed between people by as much as .5 mm., a difference that contributed significantly to variability in cell lengths and volumes (Table 9, doneby). A change in cell length due to ELF EM fields will have to be greater than this to be considered biologically relevant. However, within and between person variability is needed for a better estimate of measurement errors. A sample of cells from the 1986 nests will each be measured several times by the same person and by different people. Thus we will be able to obtain estimates of within and between person measurement error. A change in cell length or cell volume between treatment and control sites will not be considered significant unless it is greater than the sum of the within and between person measurement errors.

Sex of the offspring

The sex of the M. relativa offspring that emerged from a given cell is known only for a limited number of cells. These cells were tested with an ANOVA, treating each cell as an independent measure. Offspring's sex does not contribute significantly to the variance in either cell length or volume (Table 10), although cell order does. We will be interested to see if this result is true of the 1986 nests, and for M. inermis, which has greater sexual dimorphism in size.

Date that the nest was begun

Mean cell length and volume for a nest is not significantly affected by date that the nest was begun (Table 9). If foraging rate is lower late in the season than early, there does not appear to be any associated change in mean cell length and volume between nests. We will be interested to see if such an effect appears after the ELF antenna is operational, or if it exists for M. inermis.

Results: Number of cells per nest

Mean number of cells per nest in complete nests in bore size 4 was twice the mean number in incomplete nests (4.5 vs. 2.2). The distribution of number of cells per nest for complete and incomplete nests is given in Table 12. A chi-square test of categorical data was used to determine if exp., site [exp], or date ("early", before July 20; and "late", July 20 or later), had significantly different

distributions of cells per complete nest (bore size 4 only). None of these factors were significant (Table 13). Mean cells per nest for these different categories varied from close to 7 to just under 3 (Table 14), suggesting that it will be difficult to detect significant differences in the distribution of cells per nest for control and experimental sites, unless these differences are substantial.

Incomplete nests, by definition, do not have a full complement of reproductive cells, so we have not included them in our analysis of cells per nest. In the future, however, we may wish to compare the relative proportions of complete vs. incomplete nests in treatment vs. control sites, and early vs late season. Table 15 summarizes these proportions for the 1985 M. relativa data.

Hypothesis 2. Bees will make thicker cell caps and nest caps when exposed to ELF fields, or they will increase the proportion of nest space that is not devoted to reproduction.

Rationale

Honeybees increased the amount of propolis at their nest entrances under high voltage transmission lines, presumably in response to stress connected with electric fields at the nest entrance (Greenberg et al, 1981). This suggests the possibility that megachilid bees will respond to disturbance from ELF fields by increasing the amount of nest lining material in the bores. This may be reflected in larger cells (tested in hypothesis 1), increased cell cap length, and/or increased nest plug length. More generally, there could be an increase in the nest space that does not include cells for offspring (ie. basal and vestibular spaces, nest plugs and indentations).

This hypothesis has not yet been tested. The sample size for cell cap length is considerably smaller than the sample size for cell length because most cell caps were destroyed when the bees emerged. We will use a nested ANOVA to test for differences between experimental and control sites in cell cap length of cells in which the occupant died, and in nest plug length. Differences between treatment and control sites in proportion of non-reproductive nest space will be tested using a Chi square contingency table.

Hypothesis 3. The relative acceptability of nests oriented in a NS direction vs. nests oriented in an EW direction may change when bees are exposed to ELF fields.

Rationale

The matched directional placement of the hutches and nest openings is meant to allow us to detect preferences for nest direction in the bees. Since honeybees may use the earth's magnetic field under special circumstances to orient their comb (reviewed in Gould, 1980), it is possible that the fluctuating ELF magnetic fields could disturb any direction preference that megachilids normally have, or could cause the appearance of a preference for certain directions in order to reduce disturbance by the fields.

Comparisons of the relative acceptability of different nest orientations requires testing for independence of discrete categorical data (eg. Chi square) and generally requires a minimum of 5 nests from each hutch. Differences other than direction between pairs of hutches must be minimized for this analysis to be valid. For example, solar radiation and temperature exposures should be the same in all directions, and both hutches should be sufficiently close that resource availability is the same for bees in both hutches.

IV NEST ACTIVITY DATA

Measurement

Every year since 1983, one or more observers have gathered data on behavior of individual bees at the nest. Of particular relevance is the timing and sequence of foraging trips for different nesting materials, and the time spent in the nest manipulating these materials. In this report we focus on the collection of round pieces of leaf (LO), the collection of elongate pieces of leaf (LR, because the leaf pieces are rolled under the bee's body), and the collection of pollen (P). Usually the observer watched a single bee for several days in succession, until the nest was complete. This protocol generated a great deal of information on the variability in behavior within a bee, but less information on between-bee variability. As will be discussed in the results section, a different protocol that maximizes the number of bees observed will be recommended for future research.

Because behavior of insects is often affected by such environmental factors as temperature and wind speed, foraging trip durations could be correlated with weather conditions. Air temperature, relative humidity, solar radiation, rainfall, barometric pressure, wind direction, and wind speed have been monitored automatically with Model TI-5X instrumentation modules at one experimental and one test site. The instrumentation did not always function properly. The project recovery staff has not had time to evaluate the availability of these data, or to attempt the appropriate correlations. We will, however, discuss possible changes in environmental monitoring in our future research efforts.

Condition of the data

Five notebooks of nest activity data taken by four different observers from 1984 - 1986 have been transcribed to the computer. Three more notebooks of activity data from 1983 have not yet been analyzed. The five notebooks that we have transcribed thus far give us a good idea of how to sample nest activity next summer.

Table 16 indicates the number of bees that were observed during each year at each site, and the numbers of LO, LR, and P collecting trips that were timed at each site. Few bees were studied at any one site or in any year, although numerous individual durations were collected for most bees studied.

Hypothesis 4. The duration of a leaf-foraging trip changes when bees are exposed to ELF electromagnetic fields.

Rationale

Honeybee activity allegedly increased under high voltage electromagnetic fields (Wellenstein, 1973). If ELF fields cause a similar effect on megachilid bees, the time of leaf- and pollen-foraging trips might decrease. Alternatively, if bees become disoriented or agitated in the field, their foraging trips may increase in duration.

Leaf- foraging trips for M. inermis and M. relativa are easy to recognize behaviors on the order of a minute in duration. Many of these trips are taken in succession, so within and between bee variability can be analyzed, and a potentially large sample of leaf collecting trips can be timed.

Weather may also affect leaf and pollen collecting trip durations. Ideally, these nest activity data should be correlated with weather parameters using a canonical correlation technique to see if weather contributes significantly to variability in trip durations. If so, we will have to restrict observation periods to a suitable range of weather conditions, and/or carefully monitor these conditions and incorporate them as covariates in our analysis of trip durations so that changes in weather do not confound differences between control and experimental sites.

Results

Median durations for LO, LR, and P collecting trips, and median durations of times in the nest after these collecting trips, are given in Table 17. The distributions of durations of a given type of collecting trip (LO, LR, or P) and the durations of times in the nest after a collecting trip, are skewed, with a wide tail that may include very long durations (Figs. 10-12). We attempted to normalize these distributions with a log, a log-log, or a square root transformation. Only the LO durations were adequately normalized by a log-log transformation (Kolmogorov-Smirnov test, $P>0.15$) Transformed LO durations were tested using a "repeated testing of individuals" ANOVA model with weighted residual sum of squares (Sokal and Rohlf 1969) to minimize the effect of significant heterogeneity. No significant differences between years, time of day, or date were found for LO durations (Table 18). We believe that the small number of bees sampled and the high variability between bees has made it difficult to detect significant effects of these variables. There were not enough degrees of freedom to test for differences between experimental and control areas, or sites nested within experimental and control areas.

Using the coefficient of variation estimated in the ANOVA, we calculate that we can detect a 2.0 (or .5) fold change in the average LO duration with a power of .5 with a sample of 60 bees. A 3 (or .33) fold change in LO duration requires timing 27 bees. As can be seen in tables 3 and 4, 60 or more M. relativa nests were constructed at most sites in both 1985 and 1986, when a full compliment of 6 hutches were present. If these samples sizes persist, it should be possible to get adequate timings to detect the 2.0/.5 fold changes. For M. inermis, it may be difficult to find adequate bees to detect a 2.0/.5 fold change. Our hope is that we can increase M. inermis sample size at each control site so that at least a tripling of LO durations can be detected. We believe that even a tripling of the LO durations is worth testing for, because this reflects an increase of only 80 seconds in the LO trip duration. Such an increase might occur if the ELF EM fields cause even a small disorientation in foraging bees. Further refinement of our analysis (eg., including effects of weather) should increase our ability to detect differences between treatment and control sites.

We are continuing to search for a transformation that will normalize the other durations that have been measured, or a nonparametric test that we can use to analyze LR and P durations. It may be impossible to normalize much of these data, because bees are involved in more than one activity while they are out of the nest on foraging trips. For example, a bee leaving the nest while constructing a cell may first visit a few flowers to feed on nectar and then may cut a leaf and return to the nest. On other occasions she may spend time grooming or sunning herself before cutting a leaf for the cell. On still other occasions she may combine all three behaviors. She may cut the first leaf that she lands on, or she may test several leaves before cutting the piece that she finally brings back to the nest. The observers sitting at the nest have no way of knowing what the bee is doing while she is out of the nest, except that she returned with a piece of cut leaf. The duration of any one of these behaviors may be normally or log-normally distributed, but the sum of the durations of trips involving different combinations of these behaviors is not.

LO trips, whose durations are normalized by a log-log transformation, probably involve fewer extraneous behaviors than do LR and P trips. This is because most round leaves are collected just after the bee lays an egg, to cap the cell. The cap must be put in place rapidly to reduce the probability that a parasite will find the nest, so the bee is less likely to spend time drinking nectar or sunning before she returns with the cut leaf. However, round leaves are also cut for nest plugs. Round leaves for a nest plug do not

have to be put in the nest as hastily as round leaves in a cell cap. In our analysis we did not distinguish between durations of trips to collect round leaves for cell caps and nest plugs, because we did not anticipate that differences might exist. The information required to make this distinction is available in our original data notebooks, but is not yet on the computer. We intend to separate these two types of LO durations in the near future to see if durations of LO trips for cell caps are less variable than are durations of LO trips for nest plugs.

Some of the variability in durations of foraging trips and subsequent activity in the nest may be explained by differences in weather conditions. We have not yet had time to correlate nest activity with weather conditions. Duration of foraging trips should vary with time of day and/or time during the season if temperature affects foraging durations. No significant effects of time of day were seen in our ANOVA analysis of the LO durations. Such an effect ought to be discernable for within-bee samples, for which there are abundant data available. In contrast, LR durations may vary somewhat with time of day. Trip durations are more variable between 12:00 and 18:00, with most of the shortest durations taking place in that interval (Fig. 6).

It is clear that the protocol for measuring nest activity must change in our next field season. We will be more systematic in sampling equally over all locations, times of day and times of season than has been the case in previous years. Instead of timing a single bee for several days in succession, we will try to maximize the number of bees timed per day. Because variability in LO durations is greater between bees than within bees, we feel it would be better to time many bees on one LO trip per bee than a few bees on many LO trips per bee. In fact, since both LO durations and LR durations are rapid, and several take place in succession, it will not be difficult to time several such trips in succession for each bee. We will, however, reduce the length of time spent observing any one bee. Such data should allow us to analyze within and between bee variability in LO and LR durations much better than the current data available to us.

Because the duration of pollen collecting trips is much greater than the duration of leaf collecting trips, we will give preference to timing leaf collecting trips. If bees are disoriented by the ELF antenna, the effect is more likely to be detected for leaf collecting trips than for pollen collecting trips. A disoriented bee might be a few minutes late returning to the nest. This could increase the duration of leaf collecting trips several fold, but would have an insignificant effect on the duration of pollen collecting

trips. Furthermore, the duration of pollen collecting trips depends on the condition and abundance of flower resources, which can vary considerably from year to year. Without a good measure of resource availability, the variability of pollen collecting trips will be difficult to interpret.

While we have not yet tried to correlate trip durations with weather data, we expect to encounter problems. Ambient monitoring devices were present at only one experimental and one control site. They were not necessarily close to the hutches. Thus temperature and solar radiation may not be accurate for the hutch where nest activity was being measured. In the next field season we will take temperature readings, and note whether the nest entrance is in sun or shade, prior to timing each bee. Ambient monitoring devices will continue to provide us with rainfall, wind speed, barometric pressure and relative humidity (assuming the equipment works properly).

Hypothesis 5. The time to construct a cell or a cell cap, and the number of leaf trips per cell or cell cap change when bees are exposed to ELF fields.

Rationale

Disorientation in the field or in the nest, or the attempt to pad a cell with extra leaves, will be reflected in the time to construct a cell and number of leaf trips per cell. The construction of a cell usually takes an hour or two. Sample sizes for these parameters will be smaller than for individual trip durations, but could still be sufficiently large for analysis.

Results

There are fewer durations of cell and cell cap construction, or pollen provisioning than of the individual trips that make up these durations. These durations consist of the sum of a number of individual leaf or pollen collecting trips, as well as some trips in which nothing visible is brought back to the nest, or in which wood, sand, or other materials are brought to the nest. There are problems determining when the construction of a cap ends and the construction of a cell begins. Occasionally the observer misses the return or departure of a bee. Also, if the construction or provisioning of a cell is interrupted by nightfall, the observer may miss a trip or two after leaving for the evening, or before returning in the morning. Thus, the time to construct cells or cell caps, and the time to provision a cell can usually only be approximated, and is often somewhat underestimated.

Those times that could be estimated from the activity data are summarized in Table 19. The duration of cell cap construction was estimated from the time that the bee left the nest to collect the first LO leaf after laying its egg, until the time that the bee left the nest to collect the first LR leaf for constructing the cell. Although some of the LO leaves brought into the nest may be part of the next cell's base rather than the previous cell's cap, we could not (and thus did not) try to separate them. Only LO leaves brought into the nest during this time were included in our counts of LOs in the cell cap. The duration of cell construction was estimated from the time that the bee left the nest to collect the first LR leaf until the time that the bee left the nest to collect the first load of pollen. One to four LO leaves may be included in this cell, but were not counted among the number of LR leaves used in constructing the cell. The duration of pollen provisioning was estimated from the time that the bee left the nest to collect the first pollen load until the time that the bee left the nest to collect the first LO leaf for the cell cap. This includes the time to lay the egg. When nightfall interrupted construction or provisioning, the last time given in the evening, and the first time in the morning were used as ending and starting points respectively for each portion of the total duration.

While these data suggest some interesting differences in the way that the two species of bees partition their time and pollen resources among offspring (Table 19), the paucity of data, the long time commitment required to collect it, and the high variability, suggest that it will be very difficult to detect differences in construction and provisioning durations due to ELF EM fields. On the other hand, the number of LR leaves that are used to construct a cell may be sufficiently constant to permit detection of differences due to ELF EM fields. Rather than obtain this information from direct observation of foraging bees, we propose to count LR leaves per cell in a sample of nests at the time that they are being measured for nest architecture data. This will allow us to gather data on more cells than we can obtain by direct observation.

V EMERGENCE DATA

Hypothesis 6. Overwintering survival of megachilid bees is lower when exposed to ELF fields.

Rationale and Discussion

High voltage transmission wires lower the overwintering survival of honeybee colonies (Greenberg et al., 1981). We would like to test for a similar effect in megachilid bees. Ideally, we would like to compare control and experimental sites in the proportion of cells that suffer various sources of mortality (parasitism, death of eggs, larvae, pupae, and adults) and the proportion that molt to the adult stage and emerge. This involves examining emergence data for the nests. However, the current experimental design presents some problems with testing the hypothesis. In previous winters, nests have overwintered in Channing, rather than at field sites. Therefore, we have no baseline data about overwintering survival at the field sites. Furthermore, parasitism rates seem to be high in many nests. The hutch design creates nesting aggregations which tend to attract more parasites than would isolated nests (see Eickwort, 1973). Sites with the greatest bee populations at the nests may have proportionally greater parasitism rates. Furthermore, parasitism might increase from year to year as the nesting aggregations remain in the same place. The greater the parasitism rate, the fewer bees that are expected to emerge, or that might show overwintering mortality.

We are still considering how to overcome these problems in our future research. Since parasitism usually takes place during the summer months, any cells that yield parasites give us no information about overwintering survivorship of Megachile. Thus we tentatively propose to focus our analysis on the relative proportions of Megachile adults (including those that die in the nest, since molting takes place in the spring, and dead adults were live prepupae that survived the wintered to become adults), dead Megachile pupae (which died in the spring), dead prepupae (which died between late summer and spring), and dead larvae and eggs (which died during the summer). We will analyze data from past years to determine whether site had an effect on mortality of the larval stages that died at the site where the nest was constructed vs. prepupae, pupae and adult stages that died at Channing. In future years we will leave at least half of the nests at the site where they were constructed in order to expose the nests to ELF EM fields over the winter.

We have intentionally left out of our working hypotheses, any mention of a change in bee population levels due to ELF. While we recognize that population dynamics have been a major focus of most other ELF ecological monitoring projects, we believe that it would be a mistake to put much emphasis on the effects of ELF on overall megachilid bee populations. As mentioned in previous annual reports, and as shown in Tables 1-4 the sites differ in both quantity and composition of floral resources, and in the numbers of nests used by different species of bees. Without quantitative data on floral resources, it is impossible to explain differences in bee populations between sites. Furthermore, the availability of artificial nests will itself change the population we are trying to measure.

To avoid dealing with these problems, we feel that it is best not to view the project as a study of the effect of ELF on population changes, but rather as a study of the effects of ELF on individual behavior and individual reproductive output. Population levels at the different sites and hutches are important principally to insure that adequate sample sizes can be collected to be able to detect changes in behavior due to ELF electromagnetic fields.

VI SUMMARY

Studies of the effects of high voltage transmission lines and magnetic fields in honeybees suggest several ways that solitary megachilid bees might be affected by ELF electromagnetic fields. In particular, honeybees show greater levels of activity, reduced reproductive output, lower overwintering survival and modifications of nest structure in response to high voltage transmission lines. In addition, honeybees can detect magnetic fields and may use them in orientation. ELF EM fields may affect megachilid bees in similar ways.

Megachilid bees are particularly well suited for this study. Their investment per offspring and reproductive output per nest are easy to measure because they provide each offspring with a discrete cell, and because they readily nest in artificial nests. Three types of data have been gathered in past years: Nest architecture, nest activity, and emergence/mortality.

Two abundant species at the experimental and control sites, both in the genus Megachile, are the focus of our analysis. These species differ in size and degree of sexual dimorphism. Thus, they may be impacted differently by ELF EM fields.

Three hypotheses regarding the impact of ELF EM fields on nest architecture are being tested:

Hypothesis 1: The average size (length and volume) of cells for each offspring, and/or the average number of cells produced per nest will be altered by exposure to ELF electromagnetic fields.

Hypothesis 2. Bees will make thicker cell caps and nest caps when exposed to ELF fields, or they will increase the proportion of nest space that is not devoted to reproduction.

Hypothesis 3. The relative acceptability of nests oriented in a NS direction vs. nests oriented in an EW direction may change when bees are exposed to ELF fields.

Only 1985 data for M. relativa has been analyzed thus far. These data suggest that, prior to the ELF antenna becoming operational, there are no significant differences between experimental and control sites in cell length and volume. Fewer nest diameters will be made available in future years, in order to reduce variability and increase sample sizes of the bee species under study. A sample of 24 nests per site will be sufficient to detect a 10% change in

cell length (mean approx. 11mm) with a power off .9 and an of .05.

No significant differences between experimental and control sites were detected in number of cells per nest. However, it is difficult to estimate the number of nests required to detect a specific change in the number of cells per nest.

We have not yet analyzed the data to test hypotheses 2 and 3.

Two hypotheses regarding nest activity are being tested:

Hypothesis 4. The duration of a leaf-foraging trip changes when bees are exposed to ELF electromagnetic fields.

Hypothesis 5. The time to construct a cell or a cell cap, and the number of leaf trips per cell or cell cap change when bees are exposed to ELF fields.

The duration of round-leaf collecting trips (used in the construction of cell caps) was examined in detail. Variability between bees was greater than variability within bees. Because small numbers of bees were tested, it was impossible to test for differences between experimental and control sites or between years. However, we estimate that 27 bees per site must be timed to detect a tripling of the 40 s. leaf collecting trip with a power of .5 and an of .05. This magnitude of change is possible if bees are disoriented by ELF EM fields.

The effort required to gather dat ι the time to construct a cell or cell cap is too great be worthwhile. However, in the future we will count the num... f leaves per cell (after recording nest architecture and emergence data) to estimate number of trips required per cell.

One hypothesis concerning emergence and mortality data has not yet been analyzed:

Hypothesis 6. Overwintering survival of megachilid bees is lower when exposed to ELF fields.

In the past, all nests were overwintered at Channing, but in future years at least half of the nests will overwinter at the site where they were constructed. We are particularly interested in mortality of the prepupal stage, the stage in which the bee overwinters.

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TABLE 1: Total number of nests of the five most abundant bee species at each site. (Numbers in parenthesis indicate number of hutches with more than five nests of a given species. This should be compared with the total no. of hutches at the site.)

1983[a]

	Control Sites		Test Sites	
Site	Camp 5	County Line	Ford 1 (north)	Ford 2 (south)
No. hutches	0	2	4	4
Species				
O. tersula		NA	NA	NA
H. albifrons		NA	NA	NA
M. pugnata		NA	NA	NA
M. inermis		NA	NA	NA
M. relativa		34[b] (2)	136 (4)	24[b] (3)

[a]Data sheets are only available for M. relativa

[b]Only nests constructed late in the season were collected at these sites

TABLE 2: Total number of nests of the five most abundant bee species at each site. (Numbers in parenthesis indicate number of hutches with more than five nests of a given species. This should be compared with the total no. of hutches at the site.)

	1984[a]			
	Control Sites		Test Sites	
Site	Camp 5	County Line	Ford 1 (north)	Ford 2 (south)
No. hutches	4	4	4	4
Species				
O. tersula	7 (0)	24 (3)	17 (2)	27 (3)
H. albifrons	19 (2)	22 (2)	NA[b]	42 (3)
M. pugnata	3 (0)	10 (1)	15 (2)	42 (2)
M. inermis	6 (1)	11 (1)	90 (3)	109 (4)
M. relativa	17 (2)	30 (3)	43 (4)	44 (4)

[a]Numbers for Megachile spp. are based on unconfirmed identifications.

[b]Data not yet on the computer.

TABLE 3: Total number of nests of the five most abundant bee species at each site. (Numbers in parenthesis indicate number of hutches with more than five nests of a given species. This should be compared with the total no. of hutches at the site.)

1985

	Control Sites		Test Sites	
Site	Camp 5	County Line	Ford 1 (north)	Ford 2 (south)
No. hutches	6	6	6	6
Species				
O. tersula	13 (1)	10 (0)	7 (0)	3 (2)
H. albifrons	16 (1)	18 (1)	14 (1)	80 (5)
M. pugnata	11 (1)	10 (0)	31 (3)	113 (6)
M. inermis	32 (3)	22 (2)	232 (6)	136 (6)
M. relativa	61 (5)	87 (6)	96 (6)	132 (6)

TABLE 4: Total number of nests of the five most abundant bee species at each site. (Numbers in parenthesis indicate number of hutches with more than five nests of a given species. This should be compared with the total no. of hutches at the site.)

1986[a]

	Control Sites		Test Sites	
Site	Camp 5	County Line	Ford 1 (north)	Ford 2 (south)
No. hutches	6	6	6	6
Species				
Osmia spp.	25 (2)	20 (2)	17 (2)	16 (1)
Hoplitis spp.	11 (0)	23 (3)	6 (0)	38 (5)
M. pugnata	15 (2)	9 (0)	12 (0)	123 (6)
M. inermis	16 (2)	3 (0)	50 (3)	80 (5)
M. relativa	69 (6)	72 (6)	52 (5)	123 (6)

[a]Numbers for all species based on unconfirmed identifications.

TABLE 5: ANOVA of all cells from 1985 *M. relativa* nests.

CELL LENGTHS

Source of variation	df	SS	F	P>F
Diameter	1	70.7	76.16***	0.0001
Exp	1	3.5	0.27	0.6569
Site [exp]	2	26.3	14.15***	0.0001
Complete vs. incomplete	1	0.9	0.97	0.3246
Doneby	3	38.8	13.92***	0.0001
Date begun	1	14.2	15.29***	0.0001
Caplflg	1	49.1	52.90***	0.0001
Cell order	10	71.6	7.71***	0.0001
Cells per nest	1	5.1	5.54*	0.0188
Model	21	294.0	15.08***	0.0
Error	1035	960.7		

$\overline{X}$ = 10.6 CV = 9.1 r^2 = .23

TABLE 5: cont.

CELL VOLUMES

Source of variation	df	SS	F	P>F
Diameter	1	6739790	8063.89***	0.0
Exp	1	564	0.08	0.7996
Site [exp]	2	13479	8.06**	0.0003
Complete vs. incomplete	1	820	0.98	0.3220
Doneby	3	35857	14.30***	0.0001
Date begun	1	9916	11.86**	0.0006
Caplflg	1	33768	40.40***	0.0001
Cell Order	10	53088	6.35***	0.0001
Cells per nest	1	1434	1.72	0.1905
Model	21	7256798	413.45***	0.0
Error	1035	865052		

$\overline{X} = 301$ CV = 9.6 $r^2 = .89$

TABLE 6: Average adult live weights (mg.).

	M. inermis 1985				*M. relativa* 1983			
	N	$\bar{X}$	S.D.	CV	N	$\bar{X}$	S.D.	CV
Females	10	164.6	10.8	6.6	32	46.6	7.4	15.9
Males	15	87.6	20.0	22.8	122	34.4	7.9	22.8
Ratio F/M		1.9				1.4		

TABLE 7: Duncan's Multiple Range Test on mean cell length by cell order in the nest. Basal cell = C1.

Cell Order	N	$\bar{X}$	Duncan Grouping	
C1	277	11.0	A	
C2	217	10.6	A	B
C3	170	10.5	A	B
C6	65	10.5	A	B
C5	94	10.4	A	B
C9	14	10.4	A	B
C8	31	10.4	A	B
C7	56	10.4	A	B
C4	128	10.3	A	B
C12	1	10.2	A	B
C10	4	9.5		B

TABLE 8: Sex ratio by cell order in the nest.
Basal cell = C1

	F		M	
Cell Order	N	%	N	%
C1	24	25.3	71	74.7
C2	12	18.2	54	81.8
C3	3	6.8	41	93.2
C4	3	10.0	27	90.0
C5	2	10.0	18	90.0
C6	2	10.0	18	90.0
C7	3	18.6	13	81.3
C8	0	0.0	12	100.0
C9	0	0.0	7	100.0
C10	0	0.0	1	100.0

TABLE 9: ANOVA of mean cell length per nest weighted by 1/standard error; includes only nests in bore size 4, only cells with cap length included.

1985 *M. relativa* nests

CELL LENGTHS

Source of variation	df	SS	F	P>F
Diameter	1	3.51	4.50	0.0358
Exp	1	1.94	1.01	0.4205
Site [exp]	2	3.83	2.45	0.0900
Complete vs. incomplete	1	1.23	1.57	0.2122
Doneby	2	3.51	2.25	0.1097
Date begun	1	2.64	3.38	0.0681
Cells per nest	1	3.56	0.46	0.5010
Model	9	21.11	3.00**	0.0027
Error	136	106.25		

$\bar{X}$ = 10.7 CV = 8.24 r^2 = .166

TABLE 9 (cont.)

CELL VOLUMES

Source of variation	df	SS	F	P>F
Diameter	1	122987	252.20***	0.0001
Exp	1	1055	0.92	0.4390
Site [exp]	2	2297	2.36	0.0987
Complete vs. complete	1	917	1.88	0.1724
Doneby	2	1870	1.92	0.1509
Date begun	1	1264	2.59	0.1098
Cells per nest	1	105	0.21	0.6438
Model	9	132105	30.10***	0.0001
Error	136	66321		

$\overline{X} = 262$ CV = 8.42 $r^2 = .67$

TABLE 10: ANOVA of cells for which offspring's sex is known; bore size 4, cap length included; 1985 M. relativa nests.

CELL LENGTHS

Source of variation	df	SS	F	P>F
Diameter	1	2.00	2.07	0.1524
Exp	1	0.00	0.00	0.9844
Site [exp]	2	8.48	4.40*	0.0142
Sex	1	0.11	0.12	0.7321
Complete vs. incomplete	1	1.41	1.46	0.2295
Doneby	2	1.25	0.65	0.5224
Date begun	1	2.15	2.23	0.1381
Cells per nest	1	4.44	4.61*	0.0337
Cell order	7	23.47	2.70*	0.0065
Model	19	54.13	2.95***	0.0002
Error	130	125.42		

$\bar{X}$ = 10.8 CV = 9.1 r^2 = 0.30

TABLE 10 (cont.)

CELL VOLUMES

Source of variation	df	SS	F	P>F
Diameter	1	84873	126.84***	0.0001
Exp	1	6	0.00	0.9683
Site [exp]	2	6324	4.73*	0.0104
Sex	1	217	0.32	0.5698
Complete vs. incomplete	1	1144	1.71	0.1934
Doneby	2	966	0.72	0.4879
Date begun	1	992	1.48	0.2256
Cells per nest	1	3617	5.41*	0.0216
Cell Order	7	15102	2.51*	0.0111
Model	19	199229	15.67***	0.0001
Error	130	86990		

$\overline{X}$ = 278 CV = 9.3 r^2 = .70

TABLE 11: Distribution of number of cells per nest in complete and incomplete nests.

	Complete nests			Incomplete nests		
Bore Size	2	4	5	2	4	5
No. Cells/ Nest						
1	4	28	11	5	43	8
2	4	21	8	3	12	8
3	8	24	12		6	3
4	7	20	8		2	1
5		17	4	1	4	1
6		9	6		2	2
7	1	13	10	2	1	1
8	3	10	6		4	
9	1	9	1			
10		8				
11						
12		1				
Total	28	160	66	11	74	24

TABLE 12: Chi-Square Analysis of Variance of number of cells per complete nest; bore size 4 only.

Source of Variation	df	Chi-Square	Prob.
Intercept	10	28.36	0.0016
Exp	10	3.91	0.9511
Site [exp]	20	18.99	0.5228
Season (early vs. late)	10	3.59	0.9641
Residual	30	17.77	0.9621

TABLE 13: Mean number of cells per complete nest at experimental and control sites, early (before July 20) and late (July 20 or later) in the season (all bore sizes).

	N	$\bar{X}$	S.D.	Min.	Max.
Experimental Sites					
F1 early	24	6.96	2.56	2	12
F1 late	20	4.30	2.70	1	9
F2 early	24	3.67	2.08	1	8
F2 late	25	3.16	2.08	1	7
Control Sites					
C5 early	31	4.32	3.29	1	10
C5 late	11	4.91	2.91	1	9
CL early	18	4.44	2.77	1	10
CL late	8	2.87	1.55	1	5

TABLE 14: Number (Percent) of complete and incomplete nests by site and bore size.

	Complete		Incomplete		Total	
Site	N	%	N	%	N	%
C5	47	(78)	13	(22)	60	(16)
CL	63	(76)	20	(24)	83	(23)
F1	68	(72)	26	(28)	94	(26)
F2	78	(61)	50	(39)	128	(35)
Total	256	(70)	109	(30)	365	(100)

Number (Percent) of complete and incomplete nests by bore size.

Bore Size						
2	28	(72)	11	(28)	39	(11)
4	161	(69)	73	(31)	234	(64)
5	66	(73)	24	(27)	90	(25)
3	1		1		2	(01)
Total	256	(70)	109	(30)	365	(100)

TABLE 15: Number of bees for which nest activity data is available at each site for each year, and total number of LO, LR, and P trips timed.

SPECIES	YEAR	SITE	BEES	LO	LR	P
M. inermis	1984	CL	1	23	33	61
		F1	11	146	97	193
		F2	2	31	22	77
	1986	C5	2	43	30	26
		F1	4	49	90	114
		F2	4	76	62	66
M. relativa	1984	F1	6	136	113	221
	1985	F1	1	29	33	51
	1986	CL	5	78	12	63

TABLE 16: Median durations in seconds for LO, LR, and P trips, and times in the nest after these trips.

		M. inermis 24 bees			*M. relativa* 12 bees		
		$\overline{X}$	range	N	$\overline{X}$	range	N
LO	Out of nest	41	(24 - 78)	368	58	(32 - 116)	243
	In nest	132	(92 - 197)	363	126	(75 - 232)	247
LR	Out of nest	144	(64 - 206)	334	185	(123 - 316)	158
	In nest	174	(117 - 236)	338	170	(132 - 267)	158
P	Out of nest	1355	(938 -2057)	537	420	(274 - 824)	335
	In nest	115	(81 - 168)	550	118	(77 - 180)	337

TABLE 17: ANOVA of log log transformed LO durations for M. inermis, using MS of individual bees as an error term.

Source of variation	df	SS	F	P>F
Exp	0			
Site [exp]	0			
Time of day	1	0.76	0.58	0.4616
Time of day, squared	1	0.90	0.69	0.4219
Date	1	0.26	0.20	0.6641
Year	1	2.48	1.90	0.1932
Error	348	66.82		

$\bar{X}$ = 1.31 CV = 33.48 r^2 = .27

TABLE 18: Average number of LO, LR, or P trips and average duration for constructing a cell cap, cell lining, or provisions.

M. inermis (10 bees)

	N	# trips	CV	Duration (sec)	CV
LO trips cell caps	17	9.5 ± 4.1	43	4292 ± 3216	75
LR trips cell linings	15	14.6 ± 4.3	29	7550 ± 3203	42
P trips	16	17.4 ± 3.7	21	24315 ± 9429	39

M. relativa (7 bees)

	N	# trips	CV	Duration (sec)	CV
LO trips cell caps	13	9.1 ± 6.2	68	4693 ± 2882	61
LR trips cell linings	14	7.6 ± 0.9	12	5151 ± 2075	40
P trips	12	17.6 ± 2.3	13	10224 ± 2794	27

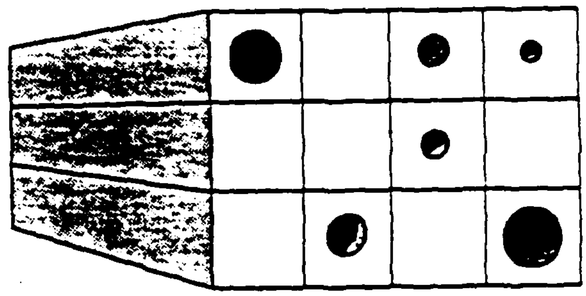


Figure 1

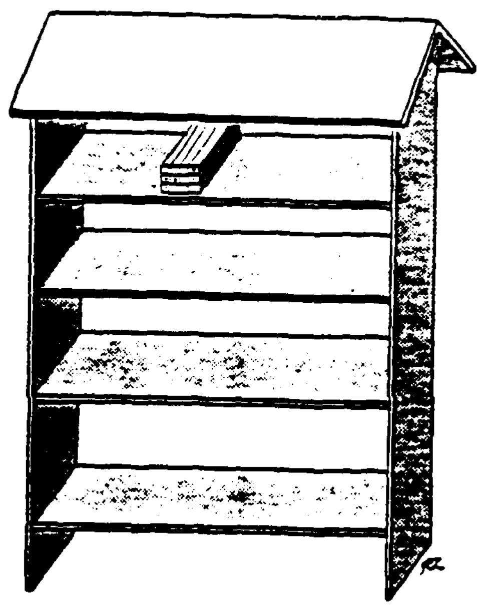


Figure 2

FIGURE 1. Nest blocks consisted of 12 trap nests, two of each of six bore sizes, each set oriented in opposite directions.

FIGURE 2. Nest blocks were placed 4 to a shelf in hutches consisting of 4 shelves. One nest block is illustrated here.

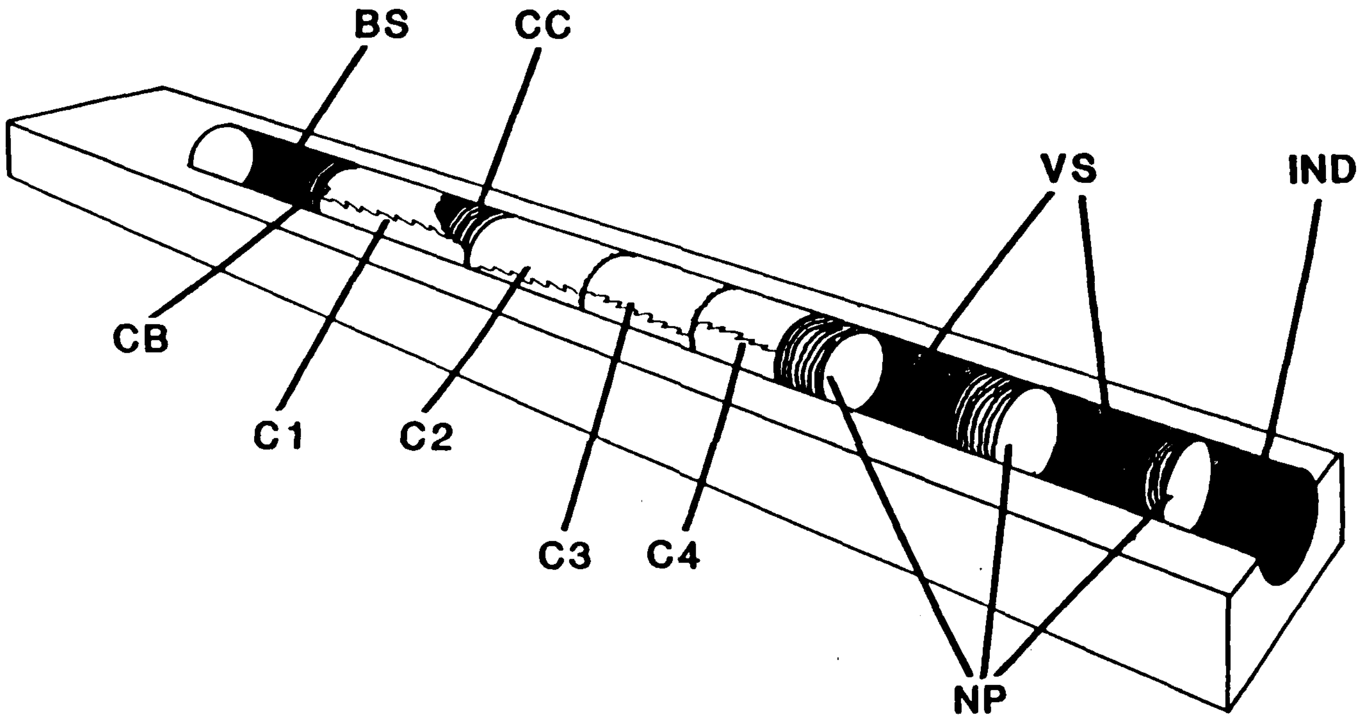
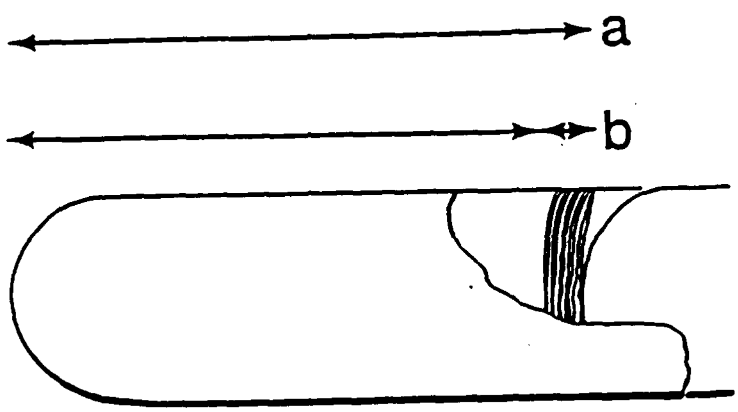


FIGURE 3. Cut away view of a completed Megachile nest.

BS - Basal Space; CB - Cell Base; C1, C2, C3, C4 - Reproductive Cells 1 through 4; CC - Cell Cap; NP - Nest Plugs; VS - Vestibular Spaces; IND - Indentation.



a – Cell Length Including Cap Length

b – Cell Length and Cap Length
Measured Separately

FIGURE 4. A single reproductive cell, indicating two ways that cell lengths were measured.

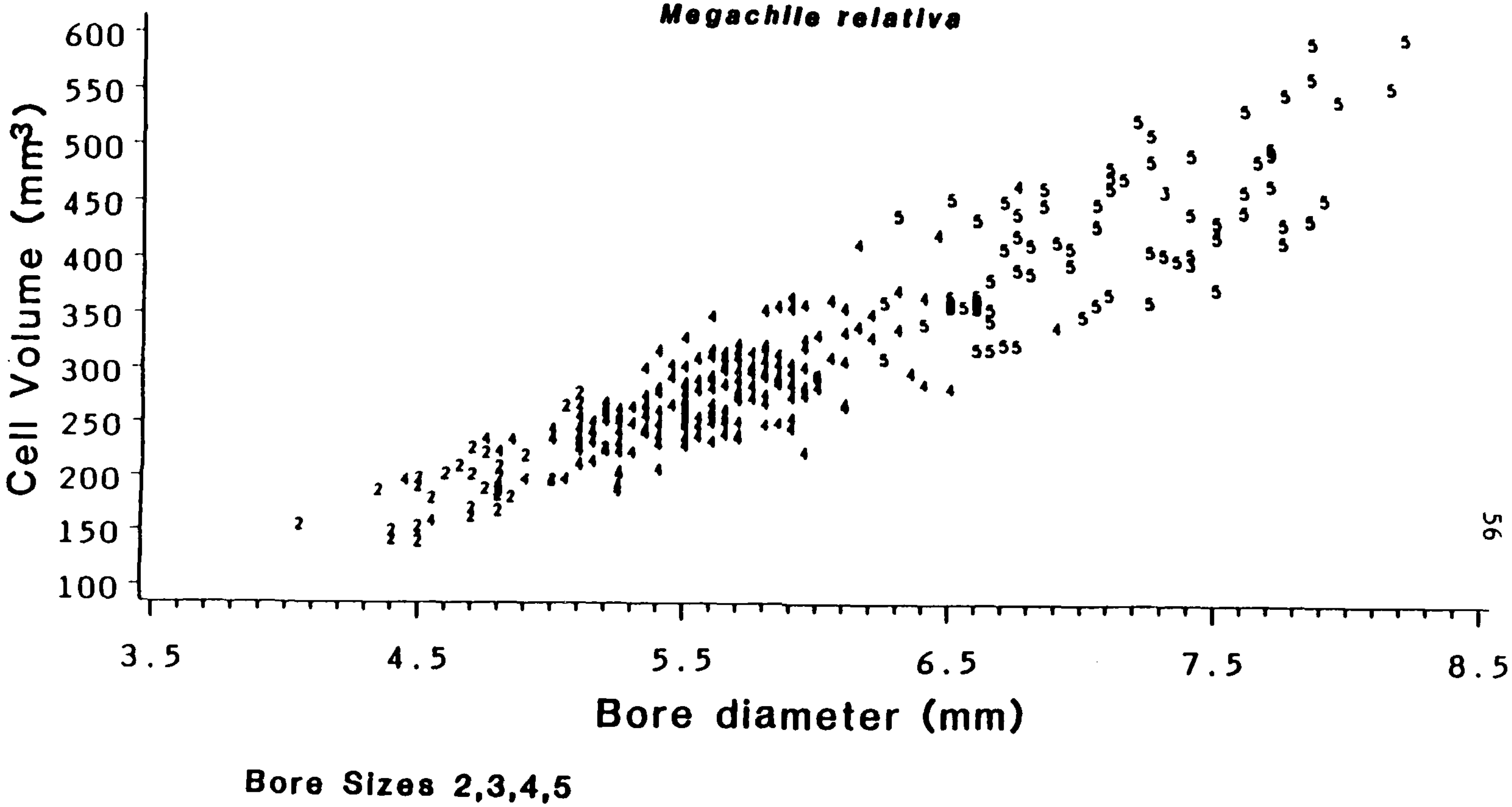


FIGURE 5. Cell volume (mm^3) versus nest diameter; Cls only, including cap volume.

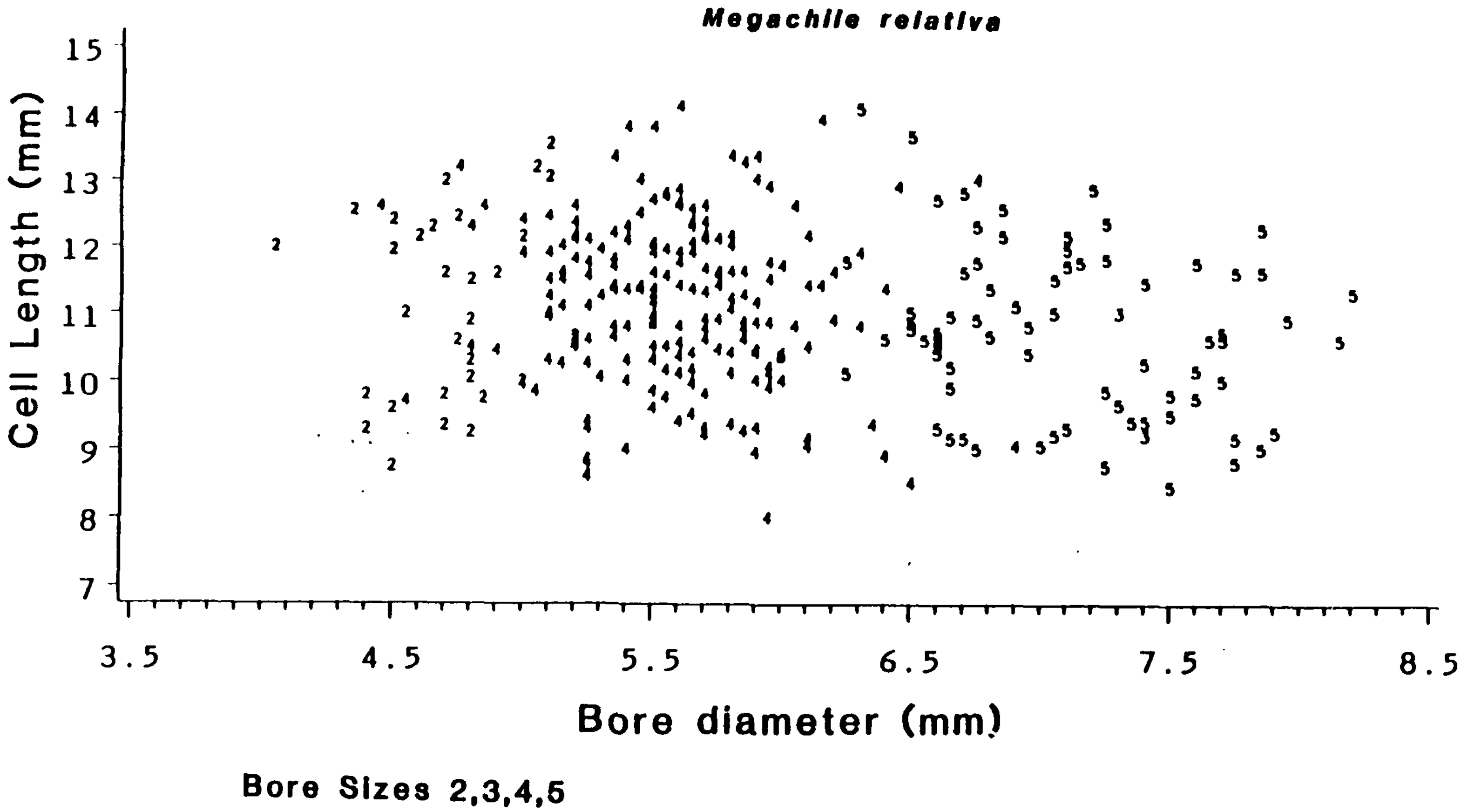


FIGURE 6. Cell length (mm) versus nest diameter; Cls only, including cap length.

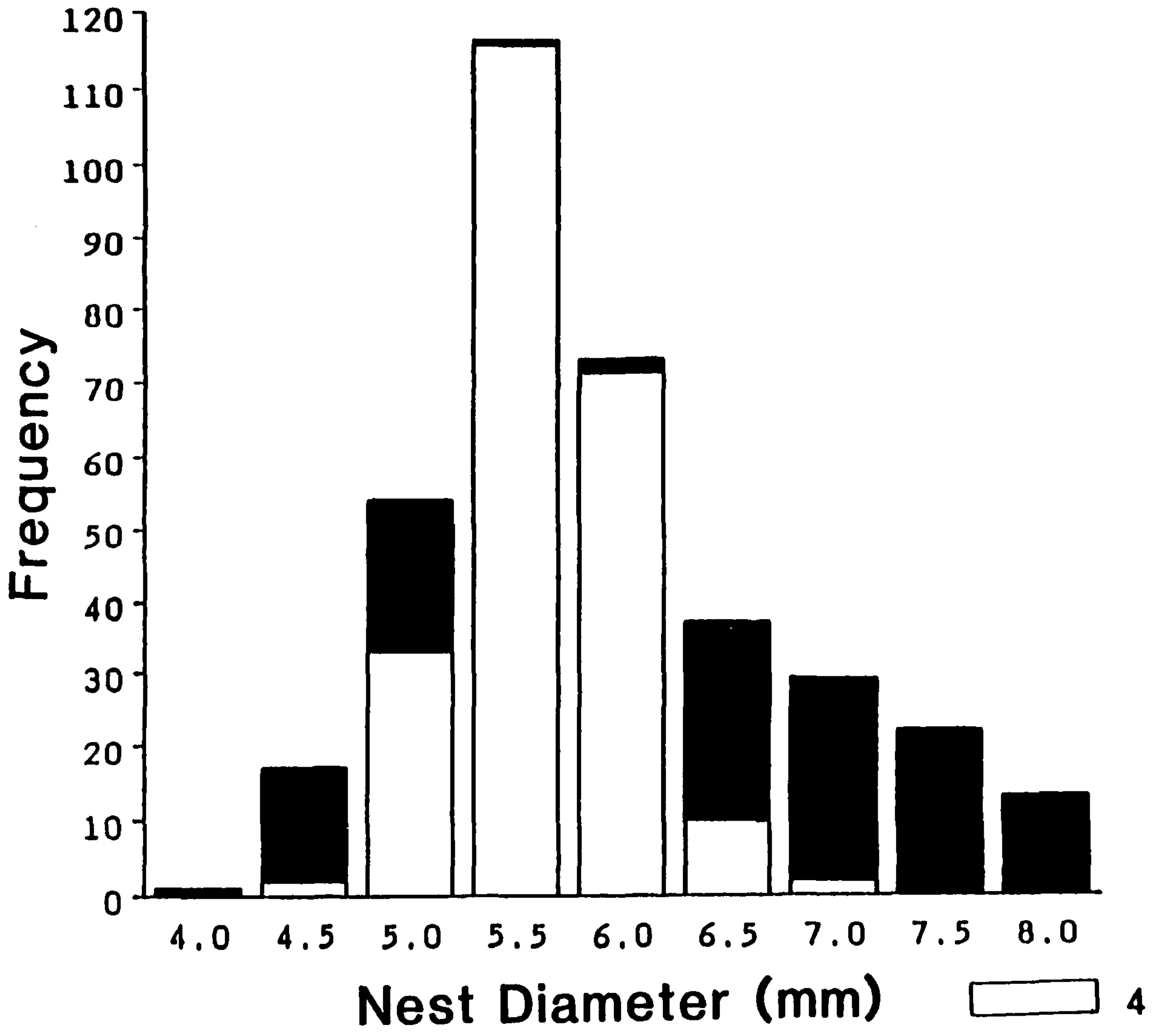
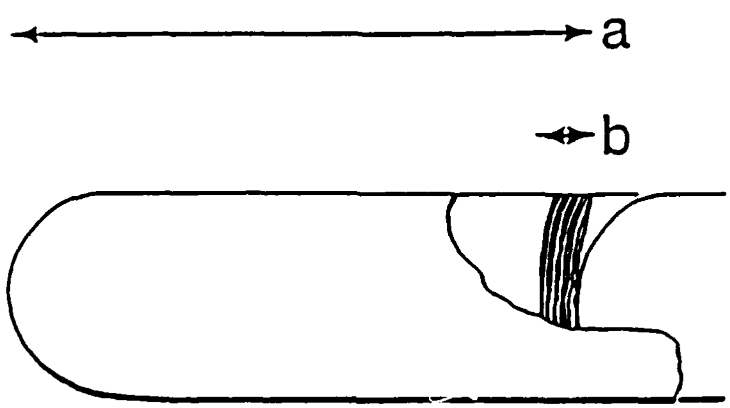


FIGURE 7. Frequencies of bore diameters for all 1985 *M. relativa* nests.



a – Cell Length Including Cap Length
b – Cap Length Measured Separately

FIGURE 8. A single reproductive cell, indicating how cell lengths will be measured in the future.

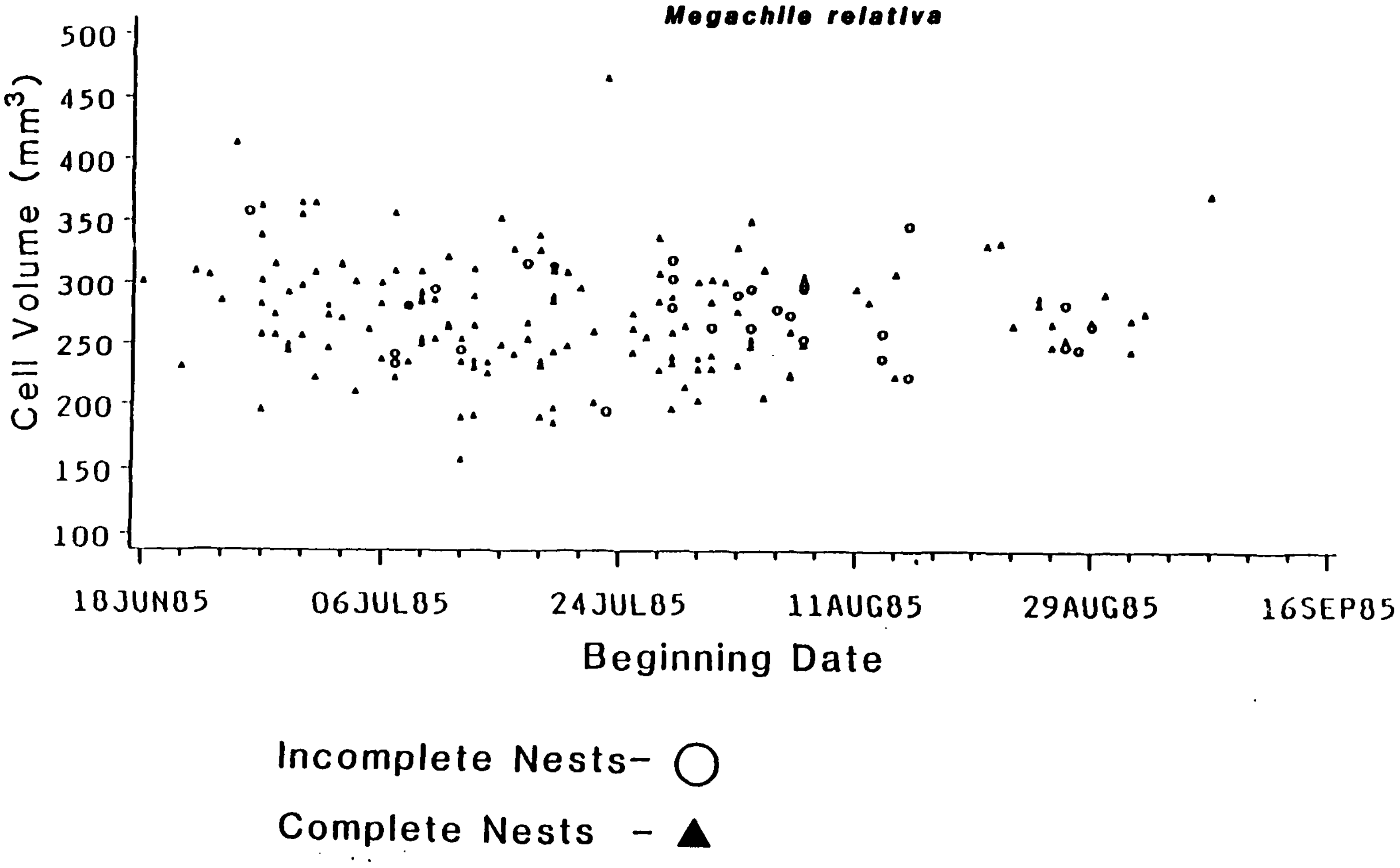
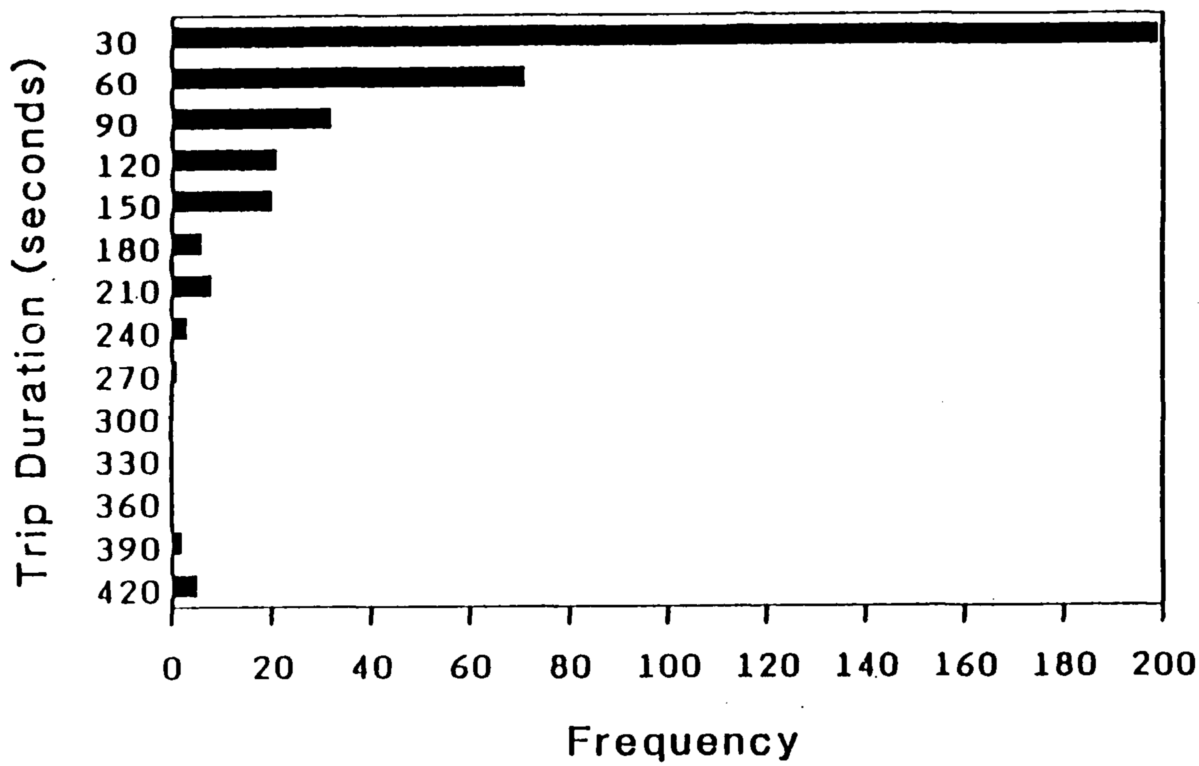
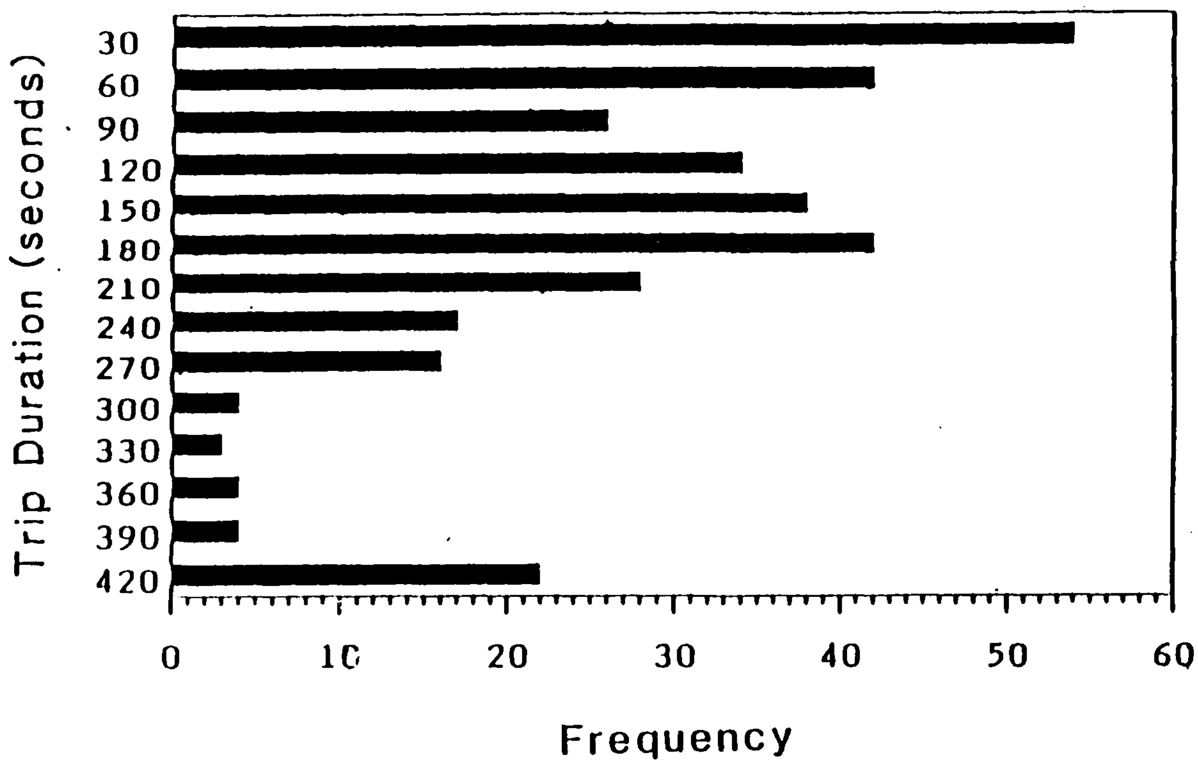


FIGURE 9. Cell length (mm) versus beginning date; Cls only including cap length; bore size 4 only.



FREQ	PERCENT
199	54.076
71	19 293
32	8.696
21	5.707
20	5.435
6	1.630
8	2.174
3	0.815
1	0.272
0	0.000
0	0.000
0	0.000
2	0.543
5	1.359

FIGURE 10. Frequency distribution of duration of round leaf (LO) collecting trips; M. inermis; data for all years and sites combined.



FREQ	PERCENT
54	16.168
42	12.575
26	7.784
34	10.180
38	11.377
42	12.575
28	8.383
17	5.090
16	4.790
4	1.198
3	0.898
4	1.198
4	1.198
22	6.587

FIGURE 11. Frequency distribution of duration of rolled leaf (LR) collecting trips; _M. inermis_; data for all years and sites combined.

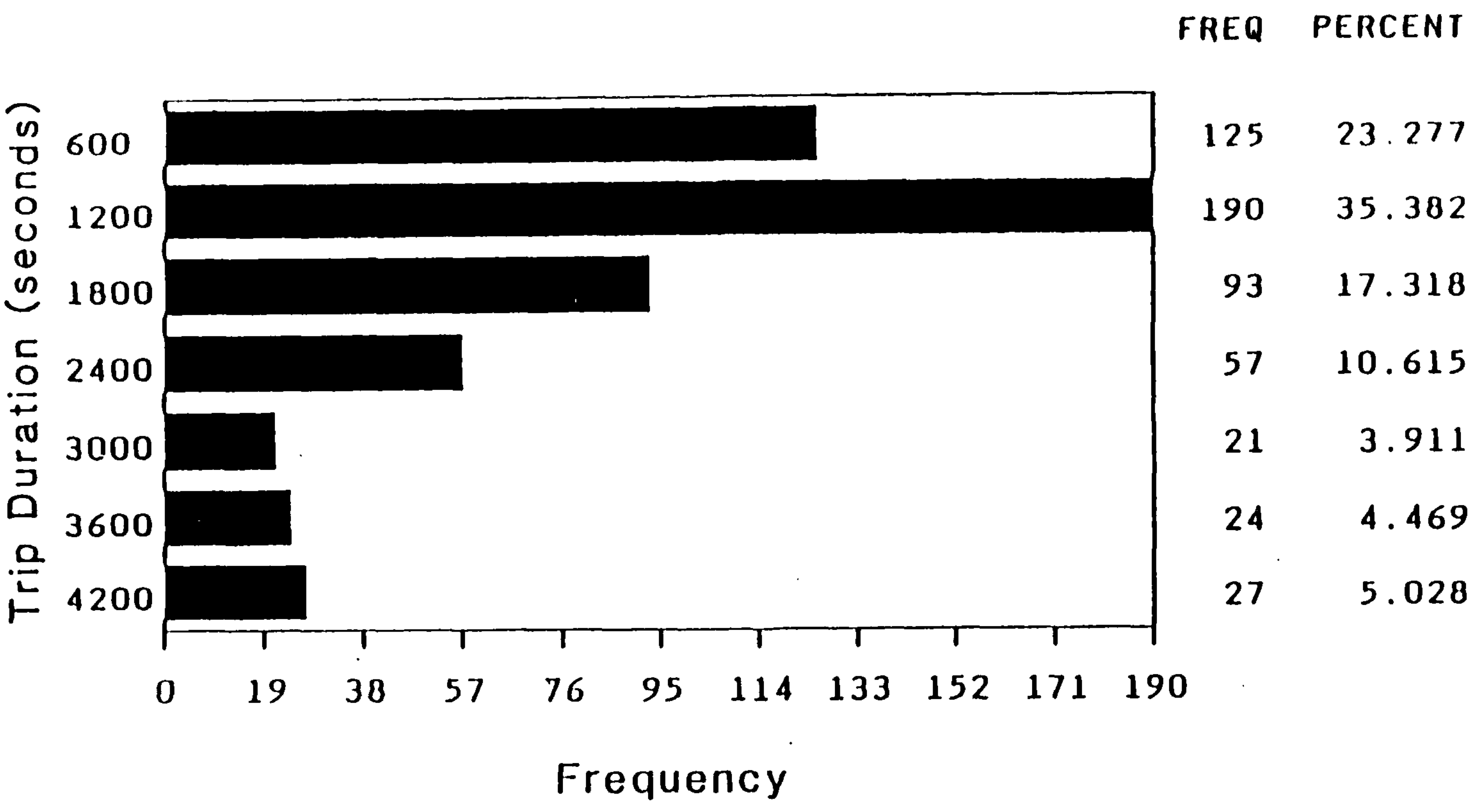
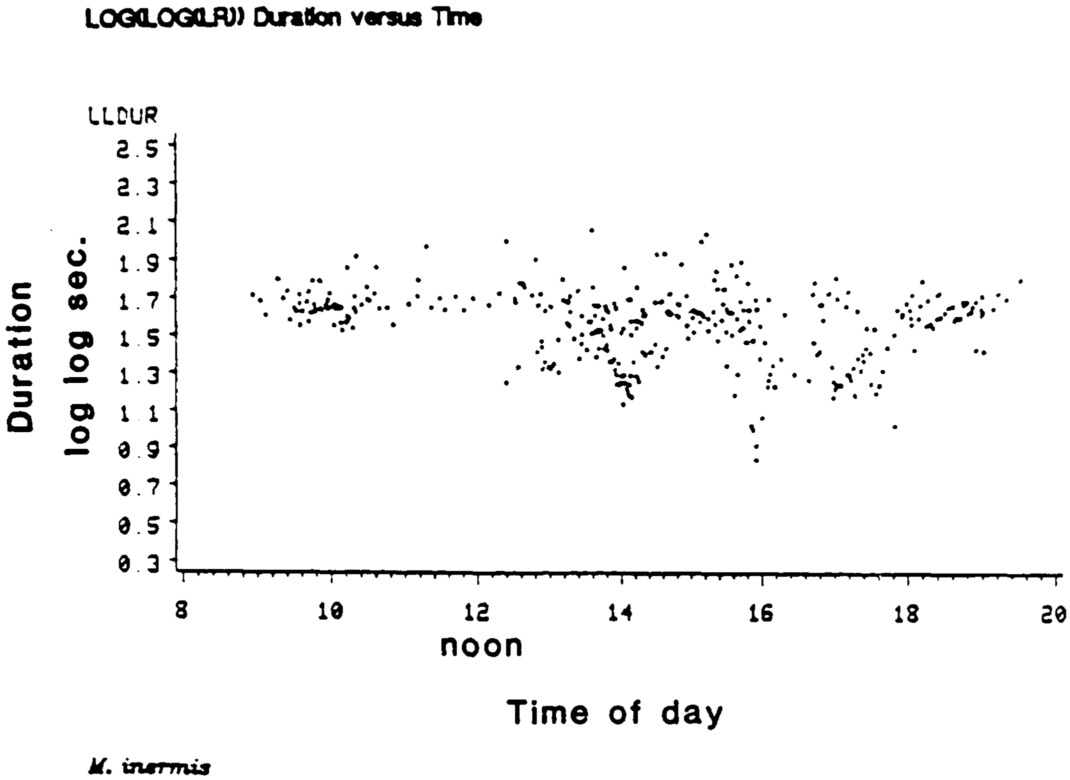


FIGURE 12. Frequency distribution of duration of pollen (P) collecting trips; M. inermis; data for all years and sites combined.



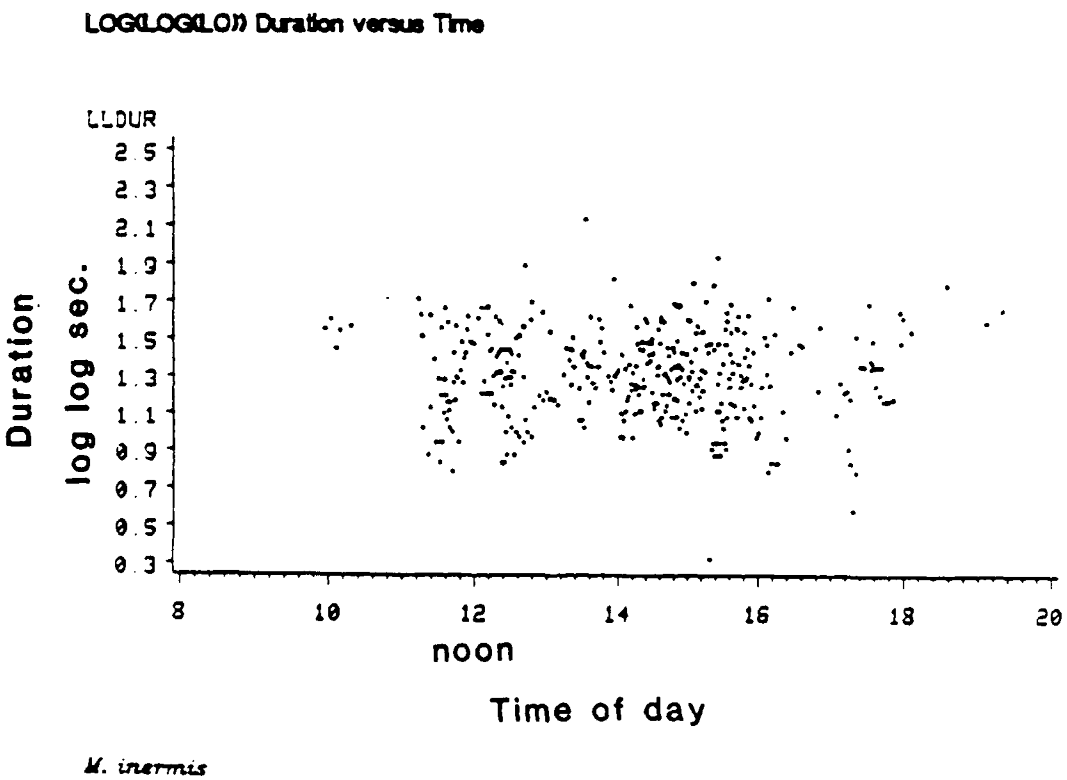


FIGURE 13. Durations of LR and LO trips plotted by time of day; data for all years and sites combined; log log transformation.

Michigan State University
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SUBCONTRACT NUMBER: E06549-84-C-006

ELF Communications System Ecological Monitoring Program

SMALL VERTEBRATES: The Michigan Study Site
Tasks 5.6, Small Mammals, and 5.12A, Nesting Birds

ANNUAL REPORT FOR 1986

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ANNUAL REPORT TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

PREFACE

This report begins with an extensive statement of the rationale for the studies proposed (see next section, titled "Rationale for Proposed Studies"). Then a section is provided on the overall research design and research facilities. Individual elements of the work are then described in detail in a series of subsequent sections. Each of the sections on individual work elements consists of three parts: (1) a brief restatement of the purpose (rationale) for the work, (2) a detailed description of research methods, and (3) a presentation of representative results gathered during prior years. The presentations of results include discussions of statistical sufficiency, including projections of the sample sizes required to discriminate between test and control plots in future years. The final section of the report summarizes the evaluation of our statistical procedures and research design that was prepared recently by an outside statistical consultant.

REPORT OF WORK ACCOMPLISHED IN 1986

Dozens of species of small birds and mammals are resident near the ELF Communication System, and the operation of the Communication System could in principle affect any of them in any of countless ways. Even with virtually unlimited resources, it would be impossible to monitor individually all ecologically important aspects of all species for possible effects of the Communication System. Accordingly, we have had to exercise informed judgement in selecting variables for study. In this process, we have been guided by two overriding goals.

Our first goal has been to monitor the overall structure of the communities of small animals. Our work in this respect is limited to mammals because the study of the structure of avian communities is the responsibility of another research group. We systematically monitor the species composition, richness, and diversity of the community of small- and medium-sized mammals, and we monitor the relative densities of two major species. By virtue of this broad-scale study of mammalian communities, we are in a position to detect diverse potential effects of the ELF Communication System on the numerous taxonomically diverse species of mammals that are resident near the System. Should the System have any sizable deleterious effects on any one or more species, many of the effects could be expected to affect measures of community richness, diversity, or relative species density, and thus we would be in a position to detect them. This is important in view of the impossibility of monitoring directly all attributes of all species.

Our second major goal has been to focus much of our effort on attributes of individual animals that are particularly likely to be susceptible to perturbation by the ELF Communication System. The reason for this focus is that laboratory research indicates that if the ELF Communication System is to have effects on birds or mammals, the effects will likely be small, and thus a statistically robust experimental design will be required to detect them (AIBS, 1985). Large numbers of independent measures can be readily obtained on individual attributes, thus facilitating statistical detection of even small effects that the ELF Communication System might have.

In our studies of attributes of individual birds and mammals, we emphasize ecologically significant variables that are especially likely to be susceptible to perturbation. Reproduction and development, for example, receive particular attention because they not only are demographically important but also are more likely to be sensitive to adverse environmental changes than many other animal properties (e.g., Goodposture, 1955; Koskimes, 1950; Kluijver, 1951; Krebs, 1970; Lack, 1954, 1966; Nice, 1954; Perrins, 1965; Perry and Rowlands, 1973). Behavior is studied in depth because it is sometimes modified readily and such modifications can have major repercussions on the lives of individuals and populations (e.g., Cohen et al., 1980; Green, 1979; Morse, 1980; O'Connor, 1978; Slobodkin, 1968).

In the following paragraphs we describe in detail the rationale for each aspect of our work on individual attributes. This work is concentrated on four particularly abundant species. The species have been carefully selected with a view to maximizing their ecological and

taxonomic diversity, so as to maximize the probability of detecting whatever diverse effects the ELF Communication System may have. The four are the tree swallow (Tachycineta bicolor), the woodland deermouse (Peromyscus maniculatus gracilis), the black-capped chickadee (Parus atricapillus) and the eastern chipmunk (Tamias striatus). To facilitate readability in the remainder of the report, they will be referred to simply as the "tree swallow", "deermouse", "chickadee" and "chipmunk", respectively.

Behavioral Studies

In view of the established sensitivity of certain types of orientational behavior to alteration by the ELF fields (e.g., Graue, 1974; Keeton et al., 1974; Larkin and Sutherland, 1977; Southern, 1969, 1971, 1972a, 1972b, 1973, 1974, 1975, 1976), orientation and homing in the tree swallow, deermouse, chipmunk, and certain other mammals are being tested to see if they are affected by the ELF Communication System. Specifically, the ability of animals to return to their home-range or territory after displacement is being assessed. We know that animals are able to find food (Krebs, 1970; Royama, 1966) and escape predators (Metzgar,1967; Watson, 1964) more effectively in their home-range or territory than in less familiar areas. Thus, any disturbance of their ability to return to their home-range or territory after wandering afar could decrease their probability of survival.

The attentive behavior of parental tree swallows and deermice is being assessed by monitoring visits to the nest containing eggs and young. Disturbance of attentive behavior by the ELF Communication System, if it occurred, could impair development of eggs or nestlings

inasmuch as the latter are dependent on parents for both food and warmth (e.g., Balen and Cove, 1972; Hill, 1972b).

Reproduction, growth, and development

The frequency and type of prenatal developmental abnormalities are examined in tree swallows (mammals are not studied in this respect because reproductive females would have to be killed to examine fetuses, and such deaths could have serious, adverse effects on population demographics). Prenatal developmental stages are especially likely to be susceptible to perturbation (Axelsson, 1954). There is, at present, no evidence to demonstrate that electric and magnetic fields of the magnitude generated by the ELF Communication System are capable of directly causing embryonic or fetal developmental defects. However, indirect effects are possible. Egg temperatures are extremely important for normal avian development. In particular, eggs must be kept warm by parental incubation. Thus should the incubation behavior of parent birds be disturbed by the ELF Communication System, developing eggs might suffer developmental abnormalities by virtue of experiencing abnormal reductions or fluctuations in temperature. (Zwilling, 1956; Hamilton, 1965).

We monitor aspects of fecundity in both tree swallows and deermice. In the birds, we count the number of eggs produced per female and the number of viable eggs and young per clutch. In the mice we monitor just numbers of young per litter. Fecundity is an important variable to study not only because it is demographically significant but also because it reflects on a number of variables that could, in principle, be affected by the ELF Communication System.

Alteration of male or female reproductive physiology could affect fecundity. Further, any serious disturbances of prenatal development in mammals or birds would likely be reflected in a decrease in fecundity inasmuch as abnormal embryos frequently fail to be born (i.e., they are resorbed in utero or fail to hatch) or are eaten or discarded by the parents soon after birth.

Postnatal mortality and the growth and development of nestling tree swallows and deermice are also followed. Any effects that the Communication System might exert on the young themselves could be reflected in altered rates of mortality, growth, or development. Alternatively, disturbances of parental attentive behavior could be influential because the rates of mortality, growth, and development of nestlings are dependent on the extent to which parents provide food and warmth (Hill, 1972b). The size of nestlings at the time of weaning or fledging is of particular interest because when young become independent of their parents, they must become substantially self-sufficient and their maturity can affect their likelihood of survival. Evidence exists that young birds that are of relatively small size at fledging are significantly less likely to survive than ones that grow to larger size while in the nest (Lack, 1966; Murphy, 1978; Perrins 1965).

Maximal aerobic metabolism

In the region of the ELF Communication System, low temperatures make winter the most physiologically stressful time of year, at least for animals such as chickadees that live wholly or predominantly above the snow. We study physiological variables that affect the ability of

chickadees and small mammals to cope with the severity of the winter climate. Deficits in the physiological ability to cope would be expected to decrease the probability of survival to the next reproductive season.

Birds and mammals keep warm in cold environments by producing heat metabolically to offset heat losses. The extent to which they can keep their body temperature above air temperature depends on how rapidly they can produce heat. In other words, the lowest air temperature at which they can maintain their usual body temperature is a function of their maximal rate of aerobic metabolism (= heat production) (Hart, 1957). In view of these principles, we measure the maximal rate of aerobic metabolism of chickadees and deermice during winter. This peak rate of heat production is informative not only because it determines the lowest air temperature at which thermoregulation is possible but also because it likely provides an index of metabolic endurance. The higher an animal's maximal rate of heat production is, the longer the animal will be able to maintain any particular submaximal rate of heat production (Astrand and Rodahl, 1977; Wickler, 1980). Endurance is important because low air temperatures demanding high heat production can persist for long periods of time.

Beyond its immediate significance for survival in a cold climate, the maximal rate of aerobic metabolism is a valuable variable to measure because it provides an index of physiological health. In fact, peak aerobic metabolism is widely used as such an index in studies of humans. In their classic Textbook of Work Physiology, Astrand and Rodahl (1977) state that "the maximal oxygen uptake is

probably the best laboratory measure of a person's physical fitness" if by fitness we mean the capacity of the individual for prolonged heavy work. Brooks and Fahey (1984), in the best of the recent texts on human exercise physiology, state that the maximal aerobic metabolism is "a good measure of fitness for life in contemporary society". Just as peak aerobic metabolism is used as an index of fitness for humans, it can also be so used in studies of animals. A deficit in the peak metabolism among individuals living near the ELF antenna would indicate that some attribute of the all-important systems involved in oxygen supply and use has been adversely affected by the ELF electromagnetic fields. Additional tests would then be required to determine the particular attribute(s) affected. The ability of the respiratory system to provide oxygen, the ability of the circulatory system to transport oxygen and nutrients to metabolically active tissues, the ability of storage tissues (e.g., adipose tissue) to mobilize stored nutrients, and the enzymatic competence of metabolically active tissues to catabolize nutrients are among the variables that influence an animal's peak rate of aerobic metabolism (Wang, 1978). In human studies, peak aerobic metabolism is usually elicited by having individuals run on a treadmill. We elicit peaks by exposing animals to cold, in part because the method is technically simpler than treadmill running (given that animals require extensive training to use a treadmill successfully) and in part because the cold-induced peak is of immediate relevance to understanding winter ecology.

OVERALL RESEARCH DESIGN AND SUPPORT FACILITIES

To detect possible effects of the ELF Communication System, we compare animal attributes on test plots (test sites) with those on paired, spatially separated control plots (control sites).

Test plots, as specified in the original IITRI Request for Proposals, are areas close enough to the Communication System that electric and magnetic fields attributable to the System, and measured in the soil near the earth's surface, will approximate 0.07 volt/meter and 0.03 Gauss, respectively. Furthermore, electric and magnetic fields attributable to ELF sources other than the System are to be at least an order of magnitude lower than those attributable to the System.

Control plots, according to the original Request for Proposals, are areas sufficiently distant from the Communication System that electric and magnetic fields attributable to the System, measured in the soil near the earth's surface, are at least an order of magnitude, and preferably two orders of magnitude, below those at paired test plots. Furthermore, electric and magnetic fields in the air and earth attributable to ELF sources other than the System are not to differ by more than an order of magnitude between the control plots and their paired test plots.

For purposes of experimental design, the test plot(s) used for any particular work element are paired with particular control plot(s). The plots of a pair are matched as closely as possible for vegetation, soil type, drainage, and other such features. By pairing plots in this

way, we minimize the likelihood that non-ELF differences between plots will introduce significant confounding effects into our results.

Different work elements are carried out on different pairs of plots for several reasons. For one thing, certain types of work could interfere with other types if both were carried out on the same populations of animals; areas where we artificially remove animals (e.g., bird embryos), for example, are not used for research on natural populations. Another factor that demands the use of different plot pairs for different work elements is that the various species we study do not all occur in similar habitat types; field habitats are required for the swallows, for example, whereas forests are required for the deermice.

Engineers provided by IITRI have measured electric and magnetic field intensities on our pairs of test and control plots, and all the pairs we now use adequately meet the standards for field intensities already described. Details of the results of the field-intensity measurements are outlined in the 1984 annual report (Beaver, et al. 1985, pp. 3-9).

To minimize potentially confounding differences between test and control plots, sham corridors have been cut through the forests at the control plots. These corridors are clearings of the same width as the corridors cut for installation of the Communication System antenna near test plots. They were cut with similar equipment, and they have been treated similarly after cutting. In brief, the sham corridors are as identical as possible to the antenna corridor except that antenna poles and wires have not been installed in the shams. Areas for animal

study on control plots and those for animal study on test plots are located about the same distance from the sham corridors and antenna corridor, respectively.

Table 1 summarizes the pairs of test and control plots used for the various work elements, and Figure 1 shows the locations of the plots. The names given to the plots in Table 1 are the standardized ones we use in all our descriptions of experiments and results. Thus, the table should be consulted if uncertainty arises concerning a plot name.

We have established the following standard of statistical sufficiency in our work. In each element of our research, we aim to gather data on a sample size that is at least large enough to give us a 90% certainty of detecting a 20% difference between test and control sites at the 5% level of significance. This is a minimal standard. Where higher standards can be met, they will be. The sample size needed to achieve at least the minimal standard can be projected once the intrinsic variability of the data is known. Research in 1984-1986 has given us information on this variability. For continuous variables, we have used the procedure in Sokal and Rohlf (1981, p. 263) to estimate sample sizes. For discontinuous variables, we have used a Chi-Square procedure described in Gill (1978, p. 82). Table 2 presents necessary sample sizes as currently projected. The estimation of sample sizes is discussed in more detail in many of the subsequent sections of this report.

Our base of operations for the on-site field and laboratory studies is a large house rented in Crystal Falls, MI (801 Crystal

Ave.). The physiology laboratory is installed there, as well as the holding facilities for temporary housing of animals used in the physiology experiments. We have a shop for construction and maintenance of field equipment and a large shed for storage of traps, cages, construction materials, and seasonal field equipment. We also have a well established data management system housed there (see below), and living space is provided for employees. We rent and maintain three pick-up trucks to provide transportation between our base of operations and field research sites in all weather conditions on a year-round basis. In addition, we rent a snowmobile and three-wheel all terrain vehicle to gain access to our more remote sites during winter and spring when travelling the entire distance by truck becomes impossible.

For data management we employ an IMS computer system. The system is multi-user and allows storage of data on fixed and removable media. Identical systems are maintained at the field laboratory in Crystal Falls and at the MSU Museum in East Lansing. Data transfer and analysis are accomplished using both systems. Field data are collected by NEC PC-8201A portable computers. We have developed software to standardize and error check field data as it is recorded. Collected data are transferred directly into the IMS system at the field laboratory each day. Transferred data are immediately edited and stored on removable and fixed disks for later analysis. Certain data are analyzed as soon as they are collected. This data management design allows us to collect and analyze large amounts of data very efficiently and accurately. The large sample sizes required in many of

our study elements necessitate the careful and accurate data handling the system provides.

Other major equipment is described in connection with individual work elements in the sections that follow.

STUDY OF SMALL MAMMAL COMMUNITIES

I. Purpose

The purpose of these studies is to characterize the mammalian communities at test and control sites and to test for possible effects of the ELF Communication System on mammalian community structure. More specifically, the following measures are compared for the two sites and for each site from year to year: species richness (S), diversity (H', which takes into account both evenness and richness), and species composition (Pielou, 1974). Relative densities of deermice and chipmunks are estimated to test for possible effects of the Communication System at the population level. These studies also provide information on the occurrence of any rare or endangered mammals at the control and test sites.

II. Methods

This year's portion of the study began on 4 August and ended 24 August. Trapping was preceded by a seven day prebait period during which trap doors were locked open. Traps were baited on the first day and then checked and rebaited as needed on the fourth day of the prebait period. The traps were unlocked and rebaited on the seventh day and checked once daily during the following two week trapping period. Longer trap periods such as this increase the chances of capturing trap shy species, and increase the accuracy of relative

density estimates used in this study (Smith et al., 1971). Each captured animal was identified to species and marked by toe-clip, furclip or fur dye to discriminate between recaptures and new individuals. Sign surveys were conducted during the prebait and trapping periods to detect the presence of species not likely to be trapped, such as deer and bear. These surveys entailed searching for and identifying feeding signs, scats, etc., of mammals at each station and between stations. Ten trap stations at 125 m intervals were situated adjacent to both the ELF right-of-way (ROW) at the test site and the sham ROW at the control site with a buffer zone of 75 m at each. One habitat type (mixed deciduous forest) was chosen in order to minimize the effects of macrohabitat differences on community parameters. Each station consisted of six small mammal Leathers live-traps and one raccoon-sized, two chipmunk-sized, and three squirrel-sized Tomahawk live-traps. All traps were positioned in suitable microhabitats within a 15 m radius of each station center. Leathers traps were supplied with polyfil bedding and baited with peanut-butter and rolled oats. Chipmunk-sized traps were set for small carnivores (e.g., weasels) and baited with beef liver or fish. Two squirrel-sized traps were baited with cracked corn (for sciurids) while the third was baited with both fish and liver (primarily for skunks). The raccoon sized trap was baited with carrots, apples, fish and liver. This regime of multiple traps per station helps eliminate bias for species specific preference for certain traps or bait types (Smith et al. 1971). In addition to the ten live-trap stations, two pitfall trap stations were set at each site to capture the smaller shrews (Sorex

spp.) which are difficult to live-trap. Each of these stations consisted of ten plastic, 4 quart containers which were set in the ground in a line with approximately 6 m spacing. Each pitfall station was situated midway between two live-trap stations. The positions of these pitfall stations is changed every year. This and the relatively small number of pitfall traps at each site (20) should minimize the effects of kill trapping on shrew populations in successive years.

Species composition, diversity and evenness are calculated from trapping data for species that are trapped, marked and released (we exclude species assessed as present based on sign). The number of animals captured for each species is the sum of all unique individuals trapped over the 14 days of trapping. Species richness is the count of species in the summed 14-day sample, species diversity (H') is calculated as H' = - SUM p_i ln p_i, and evenness is calculated as H'/ln(**s**) (following Pielou, 1975), where p_i is the proportion of the abundance of species i in the sample and **s** is the number of species. The variance of H' is calculated following Hutcheson (1970). The formulation used is a series expansion according to Bowmann et al, (1969), cited in Hutcheson (1970),

$$\mathrm{VAR}h = [\mathrm{SUM}\ p_i \ln p_i^2 - (\mathrm{SUM}\ p_i \ln p_i)^2/n + (s-1)]/2n^2 +$$

$$(-1 + \mathrm{SUM}\ p_i^{-1} - \mathrm{SUM}\ p_i^{-1} \ln p_i + \mathrm{SUM}\ p_i * \mathrm{SUM}\ p_i \ln p_i)/6n^3 + \ldots$$

where **n** is the number of all individuals of all species **s** in the sample.

A test of the diversity from two samples is also performed following Hutcheson (1970) where

$$t = \frac{H'_1 - H'_2}{(VARh_1 + VARh_2)^{1/2}}$$

with degrees of freedom

$$D.F. = [VARh_1 + VARh_2]^2 / [(VARh_1)^2/n_1 + (VARh_2)^2/n_2]$$

where n = number of individuals of all species in the sample.

Population densities are assessed using the relative measure known as Trappable Population Number (TPN). TPN values are calculated using the linear regression method commonly used in removal studies (the "Leslie method"; Giles, 1971, pp. 449-450; Smith *et al.*, 197[illegible] and 1975). Removal of trapped animals, however, is not necessary in the present study as individuals are marked when first captured, thus allowing identification of recaptures. Leaving all animals on the plot minimizes the problem of immigration of new individuals because "empty space" is not created.

III. Results - 1986

The adequacy of our sampling effort was demonstrated in prior years following the method suggested by Pielou (1974)(see Beaver, et al., 1985). Total species richness at Michigamme and Pirlot Road sites was 9 and 11, respectively. As in the past, species composition of the two communities is quite similar with most species being common and

each community dominated (with respect to number of individuals trapped) by the deermouse and chipmunk.

The Pirlot community had higher evenness and diversity (H') than Michigamme, but not significantly so (Table 3; $t = 1.208$, $P > 0.05$; t-test due to Hutcheson 1970, d.f. = 357). In 1985, the two communities were also not significantly different in H'. Bellinger's coefficient, which indicated no difference between the plots (Table 3), indicated an overall similarity of abundance rankings at the two plots. Correlation of frequency of capture at a station by species is marginally significant, and the slope of the rank of abundance of Michigamme control to Pirlot Road test is significantly different from zero ($t=3.113$, $P<0.001$) (Table 3). Therefore, there is a high degree of similarity in the small mammal fauna on the test and control plots prior to the operation of the antenna system.

The results of the TPN analyses indicate that chipmunk populations were significantly different at between plots (Table 4, $P < 0.001$), while deermouse populations were not significantly different at the two sites (t = 11.52 and 1.56, respectively; two-tailed test). Both species were much lower in abundance in 1986 than 1985. We think the lower numbers in 1986 may have been due to Tyzzer's disease. This disease appears when animals are under stress, such as may have been caused by the severe drought in the area in 1986. A number of deermice used in the growth studies were found to be suffering from this disease, and a high percentage of them died. However, populations are known to fluctuate widely from year to year in both species whether the disease is present or not. It is our expectation that between year

comparisons will be of little value in assessing ELF effects. Our data to this point do suggest comparison of abundance on plots within years will be useful.

Our adequacy of sampling community structure may be examined using the variables with variance estimates; i.e, H', regression of ranks by plot and TPN. For H' the coefficient of variation is low for test and control plots (C.V. = 6.5% and 4.7%, respectively), which will allow us to detect differences as small as 5% (Zar, 1984). Our estimates of TPN should also allow us to detect a 20% change, although there are no statistical procedures available to estimate the precise levels of difference we can expect to detect for regression parameters (Zar, 1984).

In summary, mammalian communities at both test and control sites (prior to ELF operation) are quite similar, and we should be able to detect a 20% change in community structure in terms of diversity, similarity and trappable population number of the dominant species.

PARENTAL AND NESTLING BEHAVIOR, AND FECUNDITY, GROWTH AND MATURATION STUDIES - TREE SWALLOWS

I. Purpose

The purpose of these studies is to characterize several aspects of the reproductive process in tree swallows at test and control sites and to test for possible effects of the ELF Communication System on these variables. Specifically, the following aspects of the reproductive process are compared between test and control sites and for each site from year to year: parental attentiveness to eggs and young, numbers of eggs per clutch, hatching success within clutches, rates of growth and

development of hatchlings, and nestling mortality. All of these work elements are described together in this one section because they are all carried out on the same populations of birds.

II. Methods

These studies are carried out in natural or artificial clearings where we have erected arrays of nest boxes. The boxes are made of cedar lumber and are mounted on posts, 1.5 m above the ground. Tree swallows readily elect to nest in the boxes and will tolerate considerable disturbance by investigators. The boxes can be opened to permit inspection and weighing of young. Sheets of high-density polyethylene wrapped around the posts prevent access by terrestrial predators.

When possible, adults are captured on the nest after incubation is completed and banded with U. S. Fish and Wildlife Service bands for identification. Since it has been shown that certain reproductive variables are affected by the age of the female (DeSteven 1978), most of our effort is placed on capturing females. In addition, as many young as possible are banded before fledging.

Active nests are checked daily or every other day to determine the dates that eggs are laid, how many are laid, the dates the young hatch, and overall hatching success. Monitoring of the nests for nestling growth and mortality then continues until all young have reached 16 days of age. Young tend to fledge unusually early if disturbed beyond day 16. Therefore, after day 16, nest checks are done every other day with minimal disturbance to estimate fledging success.

For studies of egg incubation, temperature-monitoring equipment is used. The tip of an EME Systems thermosensitive probe is inserted within a simulated egg, and the simulated egg is placed among the natural eggs of the clutch. The thermosensitive probe then signals when the clutch is being warmed by the parent. Data from the probe are recorded every 3 minutes, 24 hours per day, using On-site Weather Loggers made by EME Systems and NEC microcomputers. Simultaneously, the air temperature outside the nest box is monitored and recorded using a second probe.

Parental attentiveness to nestlings is monitored using video recording equipment. The male and female parents are marked for easy identification by coloring their feathers. A Canon VC200 video camera is then trained on the nest box during daylight hours, and the video images are recorded continuously using a Canon VR-30A cassette recorder. The entire system is powered by a storage battery. The recordings are later reviewed by personnel who summarize the time of day and duration of each parental visit to the nest.

For studies of growth and development, nestlings are weighed every other day with a Pesola spring scale accurate to 0.1 gm. The lengths of the tarsus, ulna, and wing (all from the right side of the body) are measured with dial calipers accurate to 0.1 mm.

Since different observers differ slightly in their techniques for measuring weights and body parts and yet it is impossible for one observer to measure all nestlings, we have multiple observers rotate among the plots so that every nestling is eventually measured by all observers. Regularly rotating the observers in this way has the effect

of submerging the variance in measurement due to observers into the error in each nestling's growth curve. This measurement protocol unfortunately prevents us from being able to block observer effects in the statistical design. However, as we show below, when we use data from each individual bird's growth curve, even the significant effects of differences in observer technique do not prevent us from being able to detect very small differences in patterns of growth.

For analysis of growth data, we use the procedure for fitting growth data to models of growth proposed by Ricklefs (1967) and used previously for tree swallows by Zach and Mayoh (1982). Briefly, the data for each nestling are subjected to curve fitting using an exponential, logistic, and Von Bertalanffy's model in a regression routine in SAS (Statistical Analysis System). The model of best fit, as judged by having the highest value of R^2, is used in subsequent analyses to obtain the rate of growth, the intercept, and the inflection point.

In past years we have detected significant differences in growth rates of young tree swallows between test and control plots. Recent evidence suggests that food availability on a plot can have a significant effect on both clutch sizes and growth rates of tree swallows (Hussell and Quinney, in press; Quinney et al., 1986). In order to determine what degree of variation between test and control plots in growth rates is the result of food resource availability, we have undertaken steps to quantify the flying insect abundance at each site. We have communicated with Dr. Hussell of the Ontario Ministry of Natural Resources and have designed a sampling scheme based on his

earlier work (see Hussell and Quinney, in press, for detailed methodology). At each tree swallow site we collect flying insects during the daylight hours in two suspended conical nets with alcohol traps. These nets are located among the nest boxes and are constructed to face passively in the wind so as to continually sample insects which either fly or are blown into the nets. Previous studies show an excellent relationship between the insects collected in this type of system and the insects delivered to young swallows in the nest by their parents (Quinney and Ankney 1985). Sampling begins before the initiation of any egg laying and ends when all young from the plot have fledged. After insects are sorted into size classes we compute an index of the biomass of flying insects determined from daily catches on each plot. This will allow us to compare the prey abundances between test and control plots and help explain differences in growth rates between plots not due to age of the adults or clutch sizes. These data will further refine our abilities to detect possible subtle differences in tree swallow reproductive measures due to electromagnetic fields associated with the Communication System.

III. Results - 1986

Tree swallow plot names, numbers of boxes at each plot, and percent occupancy for 1986 are shown in Table 5. With the placement of additional boxes on plots in the early spring we now have a full compliment of bird boxes at test and control sites. Of the 306 nest boxes monitored, 205 (67%) had egg-laying activity which is a continuation of the increase in activity observed between 1984 and 1985. This increase is due, in part, to the completed cutting of the

sham corridors around the perimeter of some of the control plots, the roller-chopping of encroaching aspen by the Michigan Department of Natural Resources, and by our efforts at predator-proofing of nest boxes. In early spring, all of the nest box poles were wrapped with a high density polyethylene to help prevent access by terrestrial predators. With increased return rates of nesting adults observed each year we have established plots which will provide adequate sample sizes for all of the tasks reported on below. Starting in 1986, we conducted all aspects of the research program on specific plots established for each individual task (see Table 1) and will continue with this protocol as originally proposed.

The age of adults breeding on the plots was quantified in earlier years by categorizing a bird as an adult if it had a high percentage of its back plumage colored iridescent green. Younger birds have mostly a gray back plumage with little green (DeSteven 1978). In 1985, we found many more young birds nesting on control than test plots (Beaver, et al 1986). In 1986, we were not able to make as complete a determination because many birds abandoned their nests due to inclement weather prior to the time we designated to assess age of adults. However, we did keep records of birds we saw on our daily visits to the plots. Less than 10% of nesting birds were young birds and there appeared to be equal numbers of them on test and control.

Summarized fecundity data for tree swallows in 1986 and comparisons to 1985 are presented in Table 6. These data were taken from the Pirlot Road test plot and Tachycineta Meadows control plot and exclude any renesting attempts. Mean clutch size in 1986 at Pirlot Road (5.3

eggs/nest) was significantly larger than at Tachycineta Meadows (4.9 eggs/nest; t-test, $P < 0.005$). Both of these values are within the range of those reported for tree swallows (Chapman 1955, DeSteven 1978, Zach and Mayoh 1982). This is continuation of the trend shown in 1985. Pirlot Road test plot has consistently had higher clutch sizes. We suspect there is more available food at Pirlot test than the controls and this could be influencing clutch size, a finding reported for tree swallows in Canada by Quinney and Hussell (in press). We should be able to examine this using the data we have on insect biomass as soon as the analysis of our insect data by Hussell is complete. Although the mean clutch sizes were significantly different, there was no difference in the distribution of clutch sizes between test and control plots during 1986 (X^2 test of independence, $X^2 = 3.3$, df = 4, $P > 0.3$).

Hatching success (Table 7) was also greater at the Pirlot Road test plot (93.4%) than at Tachycineta Meadows (84.1%) during 1986 and these differences in likelihood to hatch are marginally significant (X^2 test of independence, $X^2 = 3.99$, df = 1, $P < 0.05$). When 1986 and 1985 data are analyzed together, likelihood to hatch is shown to be independent of both plot and year ($X^2 = 4.38$, df = 3, $P > .2$) When this 4 X 2 table is broken down, into year (1985 vs. 1986 pooled over PRT and TMC) and plot (PRT vs. TMC pooled over 1985 and 1986) components, there are no detectable plot effects ($X^2 = 1.65$, $P > 0.1$), year effects ($X^2 = 0.01$, $P > 0.3$) or plot/year interactions ($X^2 = 2.716$, $P > 0.05$). The actual number of young which hatched per nest (Table 6) was not significantly different between test and control

plots during 1986 (t-test, t = 1.5, P > 0.05), and is within the range of results reported elsewhere (Low 1934, Paynter 1954).

Fledging success was slightly lower at Pirlot Road (25.4%) than at Tachycineta Meadows (27.1%) in 1986 (Table 7), but these small differences in likelihood to fledge are not significant (X^2 test of independence, $X^2 = 0.07$, df = 1, P > 0.3). When 1986 and 1985 data are analyzed together the results are highly significant ($X^2 = 40.245$, df = 3, P < 0.001), due primarily to a highly significant year effect ($X^2 = 38.14$, P > 0.001). There were no significant differences in likelihood to fledge over years between plots, nor was any significant interaction detected. The actual number of young to fledge per nest during 1986 (Table 6) was slightly higher at the Pirlot Road test plot (1.3 young/nest) than at Tachycineta Meadows control (1.2 young/nest), but not significantly higher (t-test, t = 1.5, P > 0.25). Numbers of young fledged was the lowest recorded in any years of our study, almost totally due to one episode of inclement weather which caused up to 60% mortality on some plots. The weeks preceding this cold and wet weather were very hot and dry and insect abundance was at low levels. Coupled with this lack of resource base, hatching was very synchronous in 1986 and most of the hatchlings in the nest were unable to thermoregulate adequately at the onset of the inclement weather. During this two day bad weather period very few adults were seen in the vicinity of nest boxes and many seemed to abandon the nests. Weather caused mortality has been recorded in other studies of tree swallows (Chapman 1955, Paynter 1954) and Low (1933) reports that 50% of the young he was studying died in one week of adverse weather. Even though this

severely reduced the numbers of nests and young that were available for study, we still had sufficient numbers for all of the research tasks.

The landmark events of eye opening and primary feather eruption are presented in Table 8. Mean number of days to eye opening was shorter at the Pirlot Road test plot (5.1 days) than at Tachycineta Meadows control (5.5 days), but these differences are not significant (t-test, $t = 1.95$, $P > 0.05$). Mean number of days to feather eruption was also shorter at Pirlot Road (8.7 days) than at Tachycineta Meadows (9.4 days), and this difference is significant (t-test, $t = 3.33$, $P < 0.01$). This difference could be due to food availability on the two plots. As for the case of differences in clutch size noted above, we plan to examine this possibility using the insect data we collected.

Exposure data for nests, eggs, and nestlings used to assess mortality rates was calculated using the Mayfield method (Mayfield 1961, 1975) and are presented in Table 9. Units of exposure are egg days, nestling days, and nest days. For example, one nest with five eggs observed for four days would represent 20 egg days and four nest days. Data presented here include all active nests from all plots and represent an overall nesting success analysis.

Egg mortality was significantly higher on the control plots (G test of independence, Sokal and Rohlf 1981, $G = 11.38$, $df = 1$, $P < 0.001$), while nestling mortality was significantly greater on the control plots ($G = 15.77$, $P < 0.001$). Overall nest mortality (e.g. failure of an entire nest) was not significantly different between pooled test and control plots ($G = 0.09$, $P > 0.3$), nor were there any significant differences between test and control plots when nest

mortality is partitioned between the incubation phase and the nestling phase ($G = 1.67$ and 1.05, respectively, both $P > 0.2$).

In 1986, 148 unique adults were captured and banded; 104 (70.3%) were new individuals and 44 were returns (29.7%) which were banded by us during previous seasons. This 29.7% return is an increase from 1985 when only 16.6% of adults handled were previously banded by us. In addition, as many young as possible are banded before fledging; in 1986, 271 young were banded in the nest. This number is a decrease from the 363 banded in 1985, and is a direct result of an episode of inclement weather which caused a high rate of mortality among nestlings.

According to the R^2 values obtained from curve fitting to growth data for individual birds during 1986 (Table 10), body mass, tarsus and ulna growth were best fit by the logistic model while wing growth was best fit by the exponential model.

The models which had the best fit to the data were then used to produce values for use in an analysis of variance. Within the selected growth model, nestlings whose growth curve variables (growth constant, inflection point) differed significantly from zero were subjected to a nested analysis of variance (NANOVA), with the effect of nests included within plots. The intercept was not used in the analysis since interpretation of its meaning from a biological point of view is not clear.

In general, growth rates and inflection points were most strongly affected by nests within plots and least by plot (test or control)(Tables 11 - 14). For growth constant, the only significant

plot effect was growth of body mass. For inflection point, none of the variables was found to have a significant plot effect. However, for all variables, a highly significant effect was found for nests within plots. Thus, nests differ greatly between themselves, but not between plots, for the measured variables. Table 15 presents the means and standard statistics for each variable.

In 1985, we found significant differences in the test and control plots. We attributed this difference to the age of the birds breeding on them. The control plots were new in that year, and attracted a larger proportion of young birds. Young tree swallows have been shown to have poorer reproductive success (DeSteven 1978). Therefore, if the differences in growth we have demonstrated in 1985 were due soley to age of breeding bird, we would expect to see no differences in subsequent breeding seasons. The disappearance of the plot effect in 1986 appears to confirm our expectations.

Data for patterns of incubation have not been fully analyzed at this writing. [This is because of a request by IITRI and the NAVY in late December for a complete 5 year proposal and budget for continuation of our work. The proposal was requested by 15 January, 1987, and we were able to finally complete it at the end of February, 1987.] Data were obtained on 12 nests on the test plot and 7 nests on the control plot. Representative analysis is presented for day time incubation by the female (the male does not incubate) for a single nest on each of the test and control plots. The variables examined are total time spent incubating the eggs, measured as the percentage of time on the eggs per day, the average egg temperature during

incubation, expressed as the average temperature per bout of incubation, and the average minutes of incubation per bout. Incubation does not begin abruptly in the tree swallow. Rather, the egg temperature and persistence in setting on the eggs increases to a maximum level over the first four days. Similar patterns have been reported for incubation in other species (Zerba and Morton 1983, Skutch 1962). We have selected data for our analysis from the first day the maximum egg temperature is reached, as determined by inspecting graphs of the egg temperature. Data beyond the eleventh day of incubation were not used because incubation becomes erratic just prior to the egg's hatching. We present data on six days (we do not include data on incubation at night here) of incubation for a control plot bird, and 11 days for a test plot bird (Table 17). The average percent time spent incubating the eggs is about 81% for each bird. Similar values are reported in the literature for a variety of species (Haftorn 1979; Prescott 1964; Skutch 1962, 1976; Zerba and Morton 1983a). The coefficient of variation is highest for the control bird (12.5%). The average time on the nest is not significantly different between the two birds (One-way AOV, F=0.037, P > 0.5). Mean egg temperature while incubating (Table 16) was higher for the test plot bird (One-way AOV, F=25.587, P <0.001; test done on 1/egg temp transformed data to reduce heterogeneity of variances). We do not place much significance in this result though because it is not clear how much of the difference is due to placement of the probe in the nest. We plan further tests on placing the probe in the clutch in 1987. The mean time spent (minutes) on the nest during a bout of incubation is also compared in Table 16.

The mean times are longer for the control bird, but the variance is greater than the mean for each bird, and the means are not significantly different (One-way AOV, F=1.975, P>0.20). Other workers have reported similar values for average time on the nest, and have noted the high variability in this measure (Prescott 1964; Skutch 1962, 1976; Zerba and Morton 1983b) Currently, our best variable is the percent time spent in incubation per day. Using the dispersion statistics from the two nests analyzed to date, we estimate we need to monitor 10 nests per plot to be able to detect a 20% change in incubation time.

Data for parental care of nestlings were collected in 1986 using video cameras. Our objective was to quantify the behavior of parent tree swallows at their nest following the hatching of their young. We set up video cameras on two nests. The cameras were set up early in the morning starting with the day of hatching and allowed to operate all day. The amount of time the nests were monitored ranged between 5 and 12 hours per day. Analysis of video tapes consisted of counting the number of enterings and leavings of the nestbox, and the time spent in the nest box and absent from it. Male and female at each nest were color marked and could be discerned in the video tapes. We scored their activities separately. We could also tell if foreign birds visited the nest.

We obtained the following information for each nest: 1) the number of times a bird (male or female) visited the nest in an hour, 2) the percent of the total video time that a bird spent in the nest per day, and 3) the average length of time spent in the nest per visit. These

are measures considered by other investigators studying incubation in a variety of other species (Davis, et al., Haftorn 1979, and Morton and Pereyra 1985). However, of the three variables presented in Table 17, only visits per hour will be considered because the other two measures appear to be too variable to be useful in meeting our statistical sufficiency requirements.

Two nests were studied in 1986. The data for visits per hour are presented for each sex and nest, and for each sex with the data from the two nests pooled, in Table 17.

Examined singly, females visited the nest from 6 to 7 times per hour whereas males varied from 5 to 8 visits per hour (Table 17). When visits are pooled over nests, females have slightly lower visitation rates than males, but are much less variable (Table 17). A similar study by Quinney (1986) on tree swallows in Canada revealed nest visitation rates as high as 13 trips/hr. However, he provided no statistics of dispersion so we are unable to ascertain if our data are as variable as his.

The pooled data for females yields a coefficient of variation of about 18%. We would need to monitor about 18 nests per plot to be able to detect a 20% change in visits/hour. This is about twice the number of nests we predict we can monitor with our current equipment in a single research season. Therefore, we plan to rent additional video cameras and to pool our data over years to reach our required sample size. We feel it is among our highest priorities to obtain data on this potentially important variable. Our other, more physically based, measures of potential ELF effects will allow us to detect small

changes; measures of parental care will, in our judgement, allow us to deduce cause and effect since the young depend entirely on the parents for warmth and food.

PARENTAL AND NESTLING BEHAVIOR, AND FECUNDITY, GROWTH, AND MATURATION STUDIES - DEERMICE

I. Purpose

The purpose of these studies is to characterize several aspects of the reproductive process in deermice at test and control sites and to test for possible effects of the ELF Communication System on these variables. Specifically, the following aspects of the reproductive process are compared between test and control sites and for each site from year to year: maternal attentiveness to nestlings, numbers of young born per litter, proportions of young surviving until weaning, and rates of growth and development of nestlings. All of these work elements are described together in this one section because they are all carried out on the same families of mice.

II. Methods

These studies are carried out within enclosures because free-ranging mice have been found not to remain resident in nest boxes for long enough periods for us to obtain the data desired. The enclosures are large: 6.1 by 5.8 m. Ten enclosures have been constructed within mixed deciduous forests at both the test and control plots. They are open at the top to allow free passage of atmospheric electromagnetic fields and free exposure to weather. Furthermore, they are constructed mostly of acrylic plastic sheeting, which is permeable to atmospheric electric fields according to IITRI engineers. Briefly, the walls of the enclosures consist of acrylic sheeting attached to cedar posts; the

walls project about 15 cm below ground to prevent mice from digging out, and they extend about 60 cm above ground. A 51-cm-wide sheet of acrylic is placed horizontally along the top of each wall to prevent animals from climbing over the wall. Tree trunks are sheathed with sheets of high-density polyethylene to prevent mice from climbing in or out of the enclosures via the trees. Each enclosure is provided with a nest box and a feeding and watering station. The nest box can be opened to permit access to the mice.

Small enclosures (termed holding facilities) built according to the same design, but measuring just 1.2 by 1.2 m, are also constructed at the same sites. These enclosures are used as holding facilities for mice awaiting study in the large enclosures.

The mice to be studied are captured in mixed deciduous forest near the enclosure sites. They are set up as male-female pairs. Then later the females are transferred into the large enclosures when visibly pregnant. They give birth in the enclosures and rear their young to the age of weaning.

The attentive behavior of the mother mice toward their young is monitored using treadles attached to the nest boxes. A treadle is also placed at the feeding station to monitor time spent there by the female. Treadles follow the design of Hill (1972b) and Dice (1961). Each is enclosed in a tunnel, which is positioned over the entry into the nest box or feeding station so that the mother must pass over the treadle to enter or exit. Movements of the treadle activate a mercury switch whose signals are processed in an A/D device (EME Systems). Signals from the A/D device are recorded continuously on a NEC

microcomputer, 24 hours per day. From the records, it is possible to deduce the time of each entry and exit, and thus it is possible also to compute the durations of periods spent in and out of the nest box. Because a treadle system of this sort can monitor the movements of only a single animal, the male parent cannot be present, and monitoring of the female can be carried out only until the young are about 16 days old (for at that age the young themselves start to exit and reenter the nest box).

Newborn young are toe-clipped for identification when 4 days old. From then until they are 22 days old, their growth is followed by weighing every other day to an accuracy of 0.1 g using a Pesola scale. Initial litter size and subsequent deaths are recorded. The age of eye-opening is recorded as an index of developmental rate.

III. Results - 1986

The growth and development of 7 litters from 7 females at Pirlot test plot and 9 litters from 9 females at Michigamme control plot were monitored during 1986. About half of these litters were born in early summer and the last ones in early August (Table 18) as the deermice failed to exhibit a substantial late summer reproductive peak typical of deermice in this region (Baker, 1983).

The results of growth studies are presented in Table [illegible]. [illegible] perusal of the growth in body mass of nestlings indicates that [illegible] curves often appear non-linear. Although littermates [illegible] exhibit similarly shaped growth curves, there are apparent [illegible] in curves among litters of different females as well [illegible] between litters of the same female (i.e., some [illegible]

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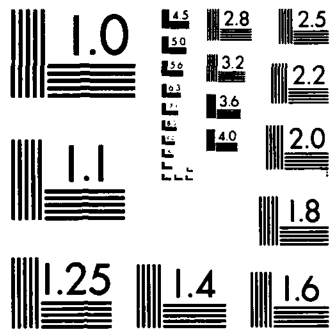

MICROCOPY RESOLUTION TEST CHART
NATIONAL BUREAU OF STANDARDS-1963-A

sigmoidal, etc.). While this variability in the shape of growth curves among (but not within litters) is interesting, it precludes the use of any particular non-linear model (e.g., logistic growth model) to estimate and compare growth rates in these mice. Therefore, growth rates have been estimated using linear regression analyses for growth of each individual, and combined growth of all individuals of each litter. For the sake of clarity, only the latter will be presented here.

The regression coefficients presented in Table 18a indicate that the linear model is adequate in describing growth of deermice (range of R^2 = .04 to .94, overall R^2 = .52). Nested ANOVA of growth rate due to mothers nested with plot yields a significant effect of mother but none due to plot (Table 18b). At this writing, we do not have any hypotheses as to the nature of the mother effect.

Litter size and age at eye opening and incisor eruption are similar between plots but exhibit large coefficients of variation (12.7% to 50%, respectively, Table 19). Much of the variation can be attributed to the frequency of visits we can make to obtain the data (now at every other day). Thus an animal categorized as not having eyes open on a particular day will not be checked again for two days. This produces a built in error of two days. Thus, we do not feel we can obtain fine enough resolution for these variables to meet our statistical criteria without increasing the frequency of visits. We are investigating this possibility within our present work schedules.

In our studies of maternal behavior, we have thus far examined the behaviors of two mothers in enclosures with treadles on both the nest

box and the feeder. The following variables were measured: (1) the number of visits to the nest or feeder, (2) the percent of time per day spent in the nest or feeder, and (3) the amount of time spent for each visit to the nest or feeder. These three variables were recorded from the time of birth of the young to 16 days after birth. We are presently in the process of analyzing these data. When the analysis is complete, we will provide IITRI and the Navy with the results.

HOMING STUDIES - TREE SWALLOWS

I. Purpose

The purpose of these studies is to measure the homing success of tree swallows at test and control sites and to test for possible effects of the ELF Communication System on such success. Variables measured are the proportions of swallows that successfully return home after displacement and the time required for each bird to return home.

II. Methods

Adult birds are captured at the nest box using a simple nest box trapping device (Cohen and Hayes 1984). Captures take place between 0930 and 1230 to allow adequate feeding of the young in the nest prior to capture. Following capture, each bird is sexed (using the presence of a cloacal protuberance for males and brood patch for females) and aged using plumage characteristics. Birds are banded using a standard U.S. Fish and Wildlife band and are color marked on the breast using "magic markers" to provide rapid and positive identification while in flight. Birds are placed in wire cages which are covered with black cloths, and then driven to the release sites.

In our first studies of swallow homing in 1984 and 1985, we released birds at all four cardinal compass directions (east, west, north, south) at test and control sites. The results revealed no differences in homing success from one compass direction to another. Furthermore, because tree swallows probably home without regard to habitats they fly over, and they are not likely exposed to any different hazards (predators, etc.) in homing from one directions as opposed to another, we feel justified in releasing birds at just one compass direction. Using just a single release point at test and control sites is more efficient in terms of personnel effort than use of four release points and thus permits adequate sample sizes to be obtained more expeditiously.

The release points are located in open areas that are at a distance of 30 km from the nest sites and at a compass direction 20 degrees NE of the nest sites (see Figure 1). This value of 30 km was chosen because it is greater than the distance corresponding to a drop of two orders of magnitude of potential electromagnetic fields given off by the Communications System. The direction of the release points in relation to the nest sites was chosen so that birds attempting to return to the test site in a straight line will cross both east-west legs of the antenna configuration -- areas that would supposedly be maximally influenced by ELF electromagnetic fields. Upon release, the time, vanishing vector, and weather conditions are noted. Observers located near the nest boxes record the time at which the birds return.

III. Results - 1986

The numbers of birds used for homing and the likelihood to return are presented in Table 20. These data are from the Panola Plains control plot and pooled from Cleveland Homestead and North Turner test plots. Test plot data were pooled after confirmation of no significant differences in return rates due to test plot location (X^2 test of independence, $X^2 = 0.09$, df = 1, $P > .3$). Likelihood to return to the nest site on the test plots was 90% (26 returned of 29 displaced) and 77% (24 of 31) on the control plots, but these differences were not significant ($X^2 = 1.62$, df = 1, $P > .2$). Mean time to return was lower on the test plots (149.8 min.) than on the control (176.9 min.), but these differences were also not significant (t-test, $t = 1.6$, $P > .05$). All of the young in the nests of birds that returned were reported in healthy condition.

HOMING STUDIES- SMALL MAMMALS

I. Purpose

The purpose of these studies is to measure the homing success of small mammals at test and control sites and to test for possible effects of the ELF Communication System on such success. Variables measured are the proportions of individuals that successfully return home after displacement and the time required for each individual to return home. The principal species studied are deermice and chipmunks.

II. Methods

The small mammal homing study is conducted on two trapping grids, one at the Pirlot road test site (PRT) and the other at the Michigamme control site (MGE). Each grid contains 100 stations spaced ten meters

apart, with one Leathers live-trap placed at each station baited with peanut butter and rolled oats. The grids were situated on the east side of both the ELF ROW (PRT) and the sham ROW (MGE). A habitat buffer between each ROW and its respective trapping grid was increased this year to 50 meters, rather than the 10 meters of 1985. This increase helped insure that both the grids and their displacement lines were located in more uniform habitat, one of continuous mixed deciduous forest dominated by sugar maple (*Acer saccharum*).

Trapping began on 6 July and ended on 23 July, 1986. Traps were checked twice daily (ca. 0800 and 1700) and rebaited as necessary. Because of the small sample sizes obtained for other species in 1985, only eastern chipmunks and woodland deer mice were displaced this year. Each animal was weighed, sexed, and toe-clipped or ear-tagged for individual identification. Reproductive condition, station number, and capture time were also recorded. Individuals were kept for displacement after their third capture; such animals were deemed to be "trap-happy" and hopefully insured their detection by continued recapture on the trapping grid upon returning from displacement. Before being displaced, each animal was kept in a laboratory cage supplied with nesting material, lab chow, and water. Cages were placed in screened-in storage sheds located near each site. Displacements took place during, or just prior to, the next activity period following capture; deermice (nocturnal) were displaced at dusk (ca. 1900) and chipmunks (diurnal) were displaced in the morning (ca. 0800). Each animal was displaced 450 m from the trap it was captured at when kept for displacement. Displacements take place to the south and west of

the home grids. The exact point of release is adjusted to reflect the point of capture on the home grid; this way all individuals are displaced exactly the same distance from their capture point. Trapping continued for five days after the last animal was displaced.

During our initial studies on mammal homing in 1985 (Beaver, et al. 1986), we displaced chipmunks and deermice in all four cardinal directions in order to investigate any directional biases in homing ability. No such biases were found even though animals displaced west and north on the control and test plots had to cross the sham corridor or actual antenna corridor, as well as somewhat different habitat types. However, our sample sizes were small for any particular displacement direction (maximum of 10 animals) and we therefore could not be certain of the robustness of our tests. Thus, in contrast to the work on swallow homing, we decided to reduce the number of displacement directions to two rather than one. Reducing the number of directions from four to two increases efficiency of sampling. By using two directions rather than one, however, we maintain the diversity of habitats and corridor crossings at each site, thus helping to insure that we are further able to examine the effects of habitat conditions as well as potential effects of ELF on homing behavior.

Displacements take place to the south and west of the home grids. The exact point of release of a displaced animal is adjusted in relation to the point of capture on the home grid so that all individuals are displaced by the same distance from their area of residence on the home grid. The home grids and the release areas are located within relatively continuous northern hardwood habitat. Once an

animal has been displaced, traps on the home grid are checked morning and evening for at least 5 days. In this way, we monitor the numbers of animals that successfully return, and we can compute the minimum amount of time required to return within about 12 hours.

The displacements to the south are through continuous forest, whereas those to the west require returning animals to cross the antenna corridor at the test site and the sham corridor at the control site. Use of the two displacement directions thus specifically allows us to test for directional differences in return rates which might occur due to the fact that animals returning from the west must pass beneath the antenna line -- potentially the area of greatest electromagnetic disturbance.

III. Results - 1986

The number of animals captured in 1986 was dramatically fewer than in 1985. The assumed reason was the presence of Tyzzer's disease in wild populations of deermice and chipmunks. We reported earlier that trappable populations of these species were down from 1985 for the presumed same reason.

A total of 9 deer mice (3 at MGE, 6 at PRT) and 41 chipmunks (22 at MGE, 19 at PRT) were displaced (Table 21) Likelihood to return to the home area was assessed using a G-test of independence (Sokal and Rohlf 1981, p. 737). No significant differences were shown between the return rates of chimpunk males and females at either the test site (PRT) or the control site (MGE) ($G = 0.203$ and 0.059, respectively, both $P > 0.3$), nor was there a significant difference between the return rates from the west and south at either site ($G = 1.438$, at

MGE; G = 0.642, at PRT; both $P > 0.2$). Also, no difference was detected in the return rate of chipmunks between the test and control sites ($G = 3.161$, $0.1 < P < 0.05$). Return rates of chipmunks for 1986 and 1985 were significantly different (adjusted $G = 7.689$, $P < 0.01$) with a higher proportion of the displaced individuals returning this year than last. This was to be expected, however, since the displacement distance for chipmunks was reduced from 1985 (500 m to 450 m). Differences in likelihood to return between years for deermice was not assessed due to the large disparity in the number of individuals displaced each year (9 in 1986, 71 in 1985). Generally, deermice are the most abundant small mammal on our forest study sites and hopefully, population numbers will increase to former normal levels. Winter trapping during 1985-1986 showed that population numbers had declined drastically since summer 1985 and these low numbers persisted throughout summer 1986, as shown by the small numbers of displacements during the homing study and the low TPN estimates during the community study. In past years, deermice have been the most abundant small mammal on our forest study sites.

DEVELOPMENTAL STUDIES

I. Purpose

The purpose of these studies is to determine the incidence of embryonic developmental abnormalities in tree swallows at test and control sites and to test for possible effects of the ELF Communication System on the incidence of these abnormalities.

II. Methods

Embryos of tree swallows are collected from test and control plots in late May and early June. Our procedure is to examine nests daily and mark new eggs. When no further eggs are laid in a nest, incubation is considered to have started. Eggs are then collected from the nest at age 96 hours (4 days) of incubation. Each embryo is dissected from the egg and placed in a fixative (Bouins solution). An initial determination of whether the embryo is normal or deformed is made at the time of dissection. At this time a determination of whether the egg was ever fertilized is made. These eggs are identified by their lack of any embryonic tissues.

The preserved specimens are later cleared, stained, mounted whole on glass slides and examined in detail for a final determination of whether they are normal or abnormal. This final determination is carried out according to a "blind" procedure. All specimens from both test and control sites are assigned arbitrary and randomly selected numbers. The person who carries out the final examination of the embryos knows only these numbers, not the origin of each specimen. Abnormal embryos are categorized according to the particular type of abnormality they show. All embryos are photographed to maintain a permanent record of normal and abnormal embryonic morphology.

III. Results - 1986

In 1986, embryos were collected from Ford North (FNT) and South test plots (FST) and Panola Plains control plot (PPC). However, in previous years as the program developed, other plots have been used. We here present data from all plots used since 1983 and compare them.

Embryos were collected from three kinds of plots: (1) a non-experimental work plot named Floodwood, (2) control plots (Tachycineta meadows (TMC) and PPC), and (3) test plots (Pirlot Road (PRT), Cleveland Homestead (CHT), FST, and FNT).

Embryos from the Floodwood plot were collected prior to the existence of control and experimental plots. Three seasons worth of data exist on embryos from this plot beginning with 1983. These embryos provided a basis for establishing normal morphologies and developmental rates and also provided a three year trend in the level of developmental abnormalities.

Embryos from the control and test plots were collected in 1985 and 1986. Because of various problems, embryos were not collected from the same sites for these two years. In future seasons, embryos will be collected from the same test and control plots to eliminate possible differences between plots within the same experimental treatment group.

The comparisons between embryos from the Floodwood plot and the control and test experimental plots reveal a significant difference in frequencies of developmental abnormalities and demonstrates that our procedures are capable of detecting differences in the frequencies of developmental abnormalities if the differences are great enough.

Floodwood: At the present time, the most extensive collection of embryos is from the Floodwood plot. Because of its location, it can not be used for a control or test plot. Table 22 presents the data for 1983 to 1985. Only one unfertilized egg was found in three years of collecting. The data appears to be homogeneous. We detected a slight but non-significant increase in the frequency of developmental

abnormalities observed in 1985. If we pool the Floodwood data over the three years, we observe a frequency of developmental abnormalities of 18.1% (Table 23). Compared to our other plots this is a very high rate of developmental abnormalities and are in the process of trying to determine why this plot should have a consistently high rate of developmental abnormalities. We note that these abnormalities cannot be a result of the ELF Communications System since it was not operative during the time the embryos were conceived or collected.

Experimental Plots: Embryos collected from control and test plots are presented in Table 24. Chi-square analysis of these data indicate that the plots are not homogeneous with respect to the frequency of developmental abnormalities. In particular one plot, Ford North Test (FNT), has an extremely high level of developmental abnormalities, although the sample size is small. The individual contribution to the Chi-square for this plot is significant by itself and renders the Chi-square significantly different at the P = 0.005 level of significance. If we eliminate the FNT data from consideration and pool the data for control and test (Table 25), we observe that the data are homogeneous and not significantly different. Thus the frequencies of developmental abnormalities observed on control and test plots, excluding the FNT data, are equal. Pooling these two sets of embryonic data together, we observed that the frequency of developmental abnormalities is about 8.0% (Table 23). We observed no infertile eggs out of 188 eggs opened. Finally, we observe that the FNT plot has a level of developmental abnormality of 34.9% (Table 23). On this plot, there was one infertile egg in 18 eggs opened.

Our data presently indicate that three different levels of developmental abnormalities exist among the plots examined. The lowest level observed was found on the experimental plots (excluding the FNT plot). If we assume that this is the normal level of abnormalities to be observed in tree swallows, we may compare this level with the levels of abnormalities observed on the Floodwood and FNT plots. Our comparison indicates that there is a significant difference between the pooled experimental plots and the Floodwood and FNT plots (Table 23). These differences are significant at least at the P = 0.005 level of significance.

The frequencies of developmental abnormalities for the pooled experimental plots and the Floodwood plot are probably reliable estimates of the levels of abnormalities that we may expect for these plots. The value for the FNT plot at this point is questionable. The sample is small and could represent a single radical fluctuation unique to the 1986 season. However, it might also mean that this plot, like Floodwood, will have a consistently high level of developmental abnormalities every year because of some intrinsic factor(s). We will examine this carefully during the next season. If the plot has a high level of abnormalities for the 1987 season, we may have to choose a different test plot to be used in our comparisons to determine the possible effects of the ELF Communication System on the frequencies of developmental abnormalities observed in tree swallows.

Developmental Retardation: Among the embryos from the 1986 season, we observed a significant number of embryos which showed developmental retardation (Table 26). That is, the embryos were

retarded in their rate of development compared to their clutch mates. The contingency Chi-square comparing these embryos, assuming that these retarded embryos are indeed abnormal, is homogeneous for all three plots (Chi-square with 2 degrees of freedom = 5.324 where the critical value at P = 0.05 is 5.991, Table 27). The overall frequency of developmental abnormalities was 26.2% (Table 27). If the FNT plot data are excluded from this comparison the frequency is 22.4% (Table 27).

At this point, we are unsure of the exact nature of these retarded embryos. Two possibilities exist: (1) they are indeed abnormal and will eventually die, or (2) they are normal and will eventually catch up with the other embryos. We may test these two possibilities by making an additional comparison. From plots not used for collecting embryos (PRT and TMC), we have hatching failure data (Tables 7 & 27). While eggs may not hatch for many reasons, if we assume all hatching failure is due to abnormalities of the embryo alone, we can compare rate of hatching failure on these plots to the rates of abnomalities on plots where eggs were collected. In many cases, the kinds of developmental abnormalities we observed should have lead to the death of the embryo in the egg and thus prevented hatching. Using homogeneous data from PPC and FST for the embryology data and homogeneous data from PRT and TMC for the hatching data (Table 27), we observe that the frequency of eggs that fail to hatch (14.4%) is intermediate between the observed frequencies of embryonic abnormalities, assuming first that the retarded embryos are abnormal (abnormality rate overall of 22.4 %) or second that the retarded embryos are normal (abnormality rate overall of 8.2%) and will

eventually catch up in development. From these results, we conclude that some of the retarded embryos may be developmentally abnormal and will die prior to hatching while others will hatch normally. It may be possible to detect these young at hatching if they have lower body mass or by if they exhibit abnormal growth rates as nestlings. At present, we have not made comparisons which would allow us to evaluate the fate of the portion of the retarded embryos which must hatch. We will make these comparisons as additional data become available.

STUDIES OF MAXIMUM AEROBIC METABOLISM

I. Purpose

The purpose of these studies is to measure the peak aerobic metabolism of animals during winter at test and control sites and to test for possible effects of the ELF Communication System on peak metabolism. The principal species studied are chickadees and deermice.

II. Methods

Collection and care of birds. To attract chickadees for study, feeding stations are established in December and kept stocked throughout the winter with sunflower seeds. Chickadees are mist netted as needed from these stations. Upon capture, birds are weighed to the nearest 0.1 g using a Pesola spring scale and marked with a colored plastic leg band for individual identification. When released from captivity, they are banded using a standard U.S. Fish and Wildlife Service band for permanent marking. Birds are housed singly in wire mesh cages (28 x 18 x 31 cm). Shelled sunflower seeds and snow or water are available ad libitum. In addition, each morning and late

afternoon, meal worms are .PA provided in excess. The cages are kept in a screened outdoor holding facility, which provides natural lighting and temperature conditions.

Collection and care of mammals. Trap shelters are established in late November, prior to any substantial snowfall. The shelters are located along wandering lines situated approximately 75-250 m from the antenna or sham corridor. Habitat is northern hardwoods dominated by maple, basswood, and elm, typical of the area. Each shelter is a plastic waste container placed upside-down on top of the ground layer, with a covered top opening which provides researcher access to the ground layer once snow is present. Mice enter the shelters through the interface between the ground layer and the wall of the shelter. One Leathers live trap is placed in the bottom of the shelter and baited with rolled oats, peanut butter, and sunflower seeds. Polyester batting is provided in the trap for nesting material. Traps are prebaited and left open one month prior to actual trapping to insure that small mammals will include the stations in their subnivean runways. Researcher travel on the sites is by snowshoe along a single trail to minimize disturbance of the subnivean air spaces which are critical to small mammal movements.

Trapping is begun at the start of January and continued intermittently — according to need for animals — through March. Work is focused primarily on the deermouse. Upon capture, individuals are toe-clipped for identification, sexed and weighed to the nearest 0.1 g with a Pesola spring scale. Once at the lab, animals are transferred to standard plastic lab cages (29 x 18 x 13 cm) with wire

lids and provided with wood shavings, polyester batting, and a diet of sunflower seeds, lab chow, and apple and snow for moisture. Cages are housed in an open outdoor facility which provides natural lighting and temperature conditions.

Laboratory methods. To elicit a peak rate of oxygen consumption, we use a refined version of the helium-oxygen (helox) method first introduced to the study of small-animal physiology by Rosenmann and Morrison (1974). Placing an animal in a helium-oxygen atmosphere at a given ambient temperature greatly increases the individual's rate of heat loss by comparison to the rate in air (mostly nitrogen-oxygen), due to the relatively much higher thermal conductivity of helox. Thus, the animal must produce heat more rapidly in helox than air if it is to maintain a stable body temperature.

Whether the rate of oxygen consumption measured in helox is in fact a true peak metabolic rate depends partly upon the ambient temperature. Identifying the true peak for an individual therefore entails studying the animal at a series of ambient temperatures. Specifically, study at a minimum of three ambient temperatures is required for a definitive determination: there should be a measurement at the temperature that elicits the peak, and also there should be measurements at temperatures higher and lower, demonstrating that the rate of oxygen consumption in helox falls off if the temperature is either raised or lowered from that eliciting the peak. Of course, the temperatures of interest are unknown at the onset of work on an individual. Thus, in principle, many measurements would have to be made on an individual before its peak would be definitively identified.

In practice, experience often permits us to know in advance the temperature at which the peak will occur. Therefore, we often need to test an animal at just three temperatures to establish its peak definitively. The spacing we have used between temperatures is 5 °C. Thus, if we test an animal in helox at three ambient temperatures that are 5 °C apart (e.g. -10, -5, 0 °C) and if the highest measured rate of oxygen consumption occurs at the middle temperature, we conclude that we have identified the animal's peak rate definitively.

Tests are not carried out on the day of capture to reduce any effect of capture stress. To further avoid adverse effects of stress, animals are tested only once on any given day.

Prior to a test animals are weighed to the nearest 0.1 g on an Ohaus triple-beam balance, and their body temperature (T_b) is measured by inserting a copper-constantan thermocouple probe 2-3 cm colonically. Then each animal is placed into a metabolic chamber. Chambers are constructed from new one-half gallon paint cans, with inflow and outflow ports in the lid. The inside surfaces are painted with 3M ECP-2200, for an emissivity of nearly 1.0. A 0.5-inch-mesh hardware cloth floor covered with Dip-It plastic coating is used to elevate the animal above the bottom of the can, thus helping to insure proper airflow around the animal and permitting urine and feces to drop away so as not to wet the animal. The outflow port of each chamber houses a 36-gauge copper-constantan thermocouple to monitor chamber temperature, which is maintained by immersion of the can in a Forma Scientific 2325 water bath using ethanol as antifreeze. All temperature probes are connected

to a Leeds and Northrup 250 Series Multipoint recorder which can be read to the nearest 0.1 °C.

Measurements are carried out during daylight hours. Food is provided during measurements. Specifically, apple is provided for the mammals, and shelled sunflower seeds and a mealworm are provided for the chickadees. The metabolism chambers for the birds are equipped with a small light that provides dim illumination; without this light, the chickadees (which are diurnal feeders) would not eat. Our decision to provide food during tests is based on extensive preliminary experimentation and is predicated on the following considerations: (1) Animals in nature are able to feed during the day; the birds are diurnal foragers, and the mammals can feed from caches. (2) In the mice, the variance in results is lower when food is provided than when it is denied. (3) In the birds, there is evidence that fasting during these types of experiments increases the probability of death.

Oxygen consumption is measured using an open-flow system. Briefly, gas (air or helox) is pumped through the metabolic chamber at a measured flow rate, and the reduction in its oxygen content is measured. From these data, the rate of oxygen use of the animal can be calculated. The oxygen content of gases is measured with an Applied Electrochemistry S3A oxygen analyzer and recorded on a Houston Superscribe potentiometric recorder. Gas flow rates are measured with Brooks 1110 rotameters. The rate of oxygen consumption is calculated according to the formulas in Hill (1972a, method B), taking cognizance of the mathematical relationship between gas composition and the output

of the S3A analyzer. We have empirically verified that the S3A analyzer reads oxygen levels in helox with the same accuracy as in air.

Animals are provided with air during an initial adjustment period (0.7-1.5 hr) and then switched to helox. Flow rates are 600 ml/min in air and 900 ml/min in helox. The adjustment period in air is terminated once the metabolic rate has remained approximately stable for 15 to 20 minutes. Upon switching to helox, a rapid transition to the new gas is made by purging the metabolic chamber at a rate of 5 liters/min for two minutes. Then the rate of flow is reduced to the 900 ml/min already mentioned. The maximal rate of oxygen consumption under the test conditions is generally achieved within 15-20 minutes after the switch to helox, and animals are rarely exposed to helox for more than 25 minutes. Following the measurement in helox, animals are quickly removed from the metabolic chamber, and a final T_b and weight are recorded.

All thermocouples have been calibrated against thermometers whose calibration is traceable to the National Bureau of Standards. Flowmeters have been calibrated against a Brooks Volumeter also having a NBS-traceable calibration.

The one aspect of the measurement procedure that is open to significant subjective judgement is the determination of the particular time interval over which the maximum oxygen consumption occurred in each experiment. Because of the subjectivity involved in this determination, a "blind" procedure will be used once the Communication System antenna has been turned on and high-resolution comparisons of test and control sites are being carried out. The relevant raw data,

as earlier noted, are recorded using a potentiometric recorder. These records are not marked as to the origin of the animals (test or control site) but instead are identified simply by arbitrary, randomly assigned numbers. The final and definitive reading of the records will be carried out by a person who knows only these arbitrary numbers.

III. Results - 1986

Results of the winter of 1986 are summarized in Table 28. None of the differences between test and control sites in either the peak rates of aerobic metabolism or weights of the animals is statistically significant according to t-tests. The peak rates of metabolism are expressed in weight-specific terms in the table. If instead we analyze the peak rates, expressed on a whole-animal basis, we again find no significant differences between test and control sites.

In Table 29, the data for the test and control sites in 1986 are pooled in the second column. The first column presents our data from 1985 (only test sites were sample that winter). The differences between the data for 1985 and the pooled data for 1986 are not statistically significant according to t-tests.

In summary, for both deermice and chickadees we have f [illegible] rates of peak aerobic catabolism do not differ either from year to or from one site to another (test vs. control).

The third column of Table 29 presents all of our data, pooled across sites and years. The values in that table represent our best estimates for the means and standard deviations for each species. Accordingly, these values have been used to project sample sizes needed to meet our minimal standard of statistical sufficiency. For deermice,

the required sample size is 6 animals per site, and for chickadees it is 5 (Sokal and Rohlf, 1981). The number of associated metabolic runs needed to establish peaks on this number of animals is well within our capabilities.

Data on peak aerobic catabolism in freshly captured animals during the winter in Michigan have been published on a closely related species of *Peromyscus* by Wickler (1980). We note that our mean result for *P. maniculatus*, 20.5 ml O_2/g*hr, is close to Wickler's mean result for *P. leucopus*, 19.5 ml O_2/g*hr.

Effects of captivity on peak metabolism. Because the determination of the peak metabolic rate of an individual requires several experimental runs spread over several days, the question arises of whether the conditions of captivity affect the peak aerobic metabolism of recently wild-caught animals. If the peak metabolism were to be rapidly influenced by captivity, a premium would have to be placed on completing our protocol as soon as possible after capture. However, if the peak remains constant for a substantial period of time, our protocol can be extended. In view of these considerations, we have undertaken extensive experiments to determine if in fact the peak metabolism of individuals changes over the course of 10-15 days in captivity.

Briefly, we have measured the peak metabolism of newly captured animals, within 4-6 days of capture, using the previously mentioned protocols, and in the process we have identified the ambient temperature at which the peak occurred. We have then held the animals in captivity and retested them, after 1 and 2 additional weeks, at the

temperature which had elicited their peak earlier. We have compared the metabolic rates measured in helox during the retestings with the original peak to determine if a change in the peak rate had occurred with time in captivity. In these experiments, the overall length of captivity (12-20 days, or more) was longer than will ever be used for routine measurements of peak metabolism but was chosen to accentuate any changes that might occur during the 5-7 days of captivity required for routine measurements.

As outlined in our 1986 annual report, results are sometimes confounded by fattening of the animals during captivity or by changes in the body temperature they maintain from one metabolic measurement to another. However, we now have collected relatively unconfounded data on 11 deermice and 8 chickadees, and the evidence is that the peak rate of aerobic catabolism does not change in either species during captivity of the length under study.

APPRAISAL OF STATISTICAL PROCEDURES

In 1986, IITRI commissioned a well-known statistician to undertake a comprehensive review of our statistical procedures as presented in our 1985-1986 annual report. The statistician proposed many small changes, most of which have already been made and are reflected in this report. His overall appraisal was positive, as reflected by his summary statement: "Overall, this report is rather well written. Clean explanations are typically given for procedures

used, including explanations for why procedures were changed [in the past]. There are some spelling and grammatical errors, but they are few and not detrimental to understanding. In general, the statistical procedures employed or proposed are appropriate and well thought out...."

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Table 1. Test-control plot pairings for the various work elements for small mammals and nesting birds. Plot code designations are those used by IITRI. They are presented here for reference.

TEST PLOTS	CONTROL PLOTS	WORK ELEMENTS CARRIED OUT
PIRLOT ROAD (1T1)	MICHIGAMME NORTH (1C3)	Small mammal enclosure studies; Small mammal community studies; Small mammal homing studies
PIRLOT ROAD (1T1)	TACHYCINETA MEADOW (1C6)	Tree swallow parental care & growth studies (on plot areas separate from other activities)
PIRLOT ROAD (1T1)	PANOLA PLAINS (1C4)	Tree swallow parental care & growth studies
CLEVELAND HOMESTEAD (1T2)	PANOLA PLAINS (1C4) TACHYCINETA MEADOW (1C6)	Tree swallow embryology & homing studies, and parental care & growth studies on separate plot areas.
NORTH TURNER ROAD (1T4)	PANOLA PLAINS (1C4)	Tree swallow homing studies.
FORD RIVER NORTH (1T5)	PANOLA PLAINS (1C4)	Tree swallow embryology studies.
FORD RIVER SOUTH (1T6)	PANOLA PLAINS (1C4)	Tree swallow embryology studies.
PIRLOT ROAD (1T1)	MICHIGAMME SOUTH (1C3)	Small mammal physiology trapping
PIRLOT ROAD (1T1)	MICHIGAMME NORTH (1C3)	Chickadee physiology trapping

Note: Cleveland Homestead, Ford River North and South plots are small. This is why we designate them all as tree swallow embryology study sites.

Table 2. Minimum sample size requirements estimated for various study elements to meet the statistical standard of 90% certainty of detecting a 20% change at the 5% level of significance. The procedure follows Sokal and Rohlf (1981, pg 247) for parametric statistics and Gill (1978, pg 82) for frequencies.

STUDY ELEMENT	SPECIES	VARIABLE	ESTIMATED N / PLOT
Parental care, fecundity, growth, and maturation	deermice	litter size	11 females
		weight	21 individuals
		age eye open	6 individuals
		homing likelihood	59 individuals[ab]
		% time in nest	35 females[b]
	Chipmunk	homing likelihood	44 individuals[a]
	tree swallow	clutch size	23 nests
		egg weight	17 eggs
		likelihood to hatch	44 eggs
		mean hatch rate	52 eggs[a]
		growth rate:	
		weight	58 nestlings
		tarsus	38 nestlings
		ulna	27 nestlings
		wing	6 nestlings
		feather eruption	7 nestlings
		age eye open	37 nestlings
		fledging rate	337 nestlings[b]
		likelihood to fledge	58 fledglings[a]
		time to fledge	14 fledglings
		homing times	67 birds[b]
		likelihood to home	46 birds[a]
		% time incubating	10 nests
		N nest visits/hour	18 nests[b]
Developmental Abnormalities	tree swallow	frequency of normal embryos	48 embryos[a]
Physiology	deermice	peak metabolism	6 individuals
	black-capped chickadee	peak metabolism	5 individuals

[a] Estimated using contingency table procedure in Gill (1978).

[b] We consider these sample sizes unobtainable in a single year. However, we expect to be able to pool data across years and thereby meet the established standards.

Table 3. Summary of community variables for forest sites at Michigamme (MGE = control) and Pirlot Road (PRT = test) during 1986.

	MGE	PRT
DIVERSITY MEASURES[a]		
Number of unique individuals	182	179
Total Species Richness (S)	10	12
S used to calculate H'	8	10
Diversity : H' (Variance)	1.332 (0.0057)	1.468 (0.0070)
Evenness : E (max = 1.00)	0.641	0.638
Statistics :	$t = 1.208$ ($P > 0.20$) d.f. = 357 (2-tailed)	
SIMILARITY MEASURES[b]		
Bellinger's Coefficient (BC based on frequency at which species are more abundant in community one vs. two; χ^2	BC = 0.33^{ns} (vs. 3.84)	
RANK CORRELATION OF GENERAL ACTIVITY[c]		
Spearman's	$r = 0.53$ ($P \leq 0.0001$)	
Linear Regression	(Rank MGE) = 0.714 (Rank PRT) SE of slope = 0.229 (df = 11)	

[a] H' = Shannon-Wiener diversity calculated using Pielou's (1975) method; Variance calculated following Hutcheson (1970).

[b] Note: BC = $(M - P)^2/(M+P)$, where M and P = the number of species which are more abundant at Michigamme or Pirlot, respectively.

[c] Ranks = number of stations with this species; total of 12 species.

Table 4. Estimates of trappable population number (TPN) of eastern chipmunks and deermice at Michigamme (MGE) and Pirlot Road (PRT) forest sites during 1985 and 1986. [a]

		MGE			PRT		
Species	Year	TPN	(SE)	R^2	TPN	(SE)	R^2
chipmunks	1985	92.46	(9.129)	.69	61.82	(3.954)	.74
	1986	51.62	(7.313)	.48	23.48	(5.207)	.28
deermice	1985	144.28	(16.15)	.62	152.54	(21.22)	.63
	1986	112.38	(15.69)	.44	115.34	(14.47)	.52

[a] TPN was estimated using the Leslie method where TPN = intercept of the curve described by $CI = b_c + b_1 (NI^{.5})$, CI = the cumulative number of individuals captured to date, and NI = number of number of new individuals captured each day (N = 14 days; see text). Estimates of the intercept of this relationship were taken from the results of linear regression analyses of the transformed data (i.e., CI as a function of $NI^{.5}$).

Table 5. Tree swallow plots, number of boxes, and percent with egg laying activity for on test and control plots for 1986. Egg laying activity is defined as at least two eggs laid before abandonment or continuation of nesting.

PLOT NAME		NUMBER OF BOXES	% ACTIVITY
CLEVELAND HOMESTEAD TEST		37	62
NORTH TURNER TEST		47	60
FORD NORTH TEST		17	47
FORD SOUTH TEST		20	55
PIRLOT ROAD TEST		36	72
PANOLA PLAINS CONTROL		75	77
TACHYCINETA MEADOWS CONTROL		74	69
TOTALS	TEST PLOTS	157	61
	CONTROL PLOTS	149	73

Table 6. Tree swallow fecundity data compared for 1985 and 1986. Data are from the Pirlot Road test plot and Tachycineta Meadows control plot and excludes any renests which may have occurred.

	CLUTCH SIZE*			HATCH RATE**			FLEDGE RATE***		
	$\bar{X}$	SD	n	$\bar{X}$	SD	n	$\bar{X}$	SD	n
TEST 1986	5.3	0.88	23	5.1	1.54	14	1.3	2.27	14
CONTROL 1986	4.9	1.07	48	4.4	1.34	30	1.2	2.00	27
TEST 1985	5.4	0.87	21	4.4	1.12	11	3.6	0.84	10
CONTROL 1985	4.8	0.86	19	4.3	1.06	10	2.6	1.90	7

* Clutch size is the maximum number of eggs layed in a nest.

** Hatch rate is the number of eggs which hatch of those available to hatch -- not always the maximum number of eggs in the nest due to occasional predation.

*** Fledge rate is the number of young that fledge from the eggs which hatch, and only include those nests which were followed to completion.

Table 7. Likelihood to hatch and fledge for tree swallows in 1985 and 1986. Data are from the Pirlot Road test plot and Tachycineta Meadows control plot. Comparisons were made using a X^2 test of independence (df=3).

** HATCHING SUCCESS **

	HATCH	NOT HATCH	TOTAL	
TEST 1986	71	5	76	93.4%
CONTROL 1986	132	25	157	84.1%
TEST 1985	48	8	56	85.7%
CONTROL 1985	43	5	48	89.6%

$X^2 = 4.38 \quad P > 0.1$

** FLEDGING SUCCESS **

	FLEDGE	NOT FLEDGE	TOTAL	
TEST 1986	18	53	71	25.4%
CONTROL 1985	32	86	118	27.1%
TEST 1985	32	11	43	74.4%
CONTROL 1985	18	13	31	58.1%

$X^2 = 40.25 \quad P < 0.001$

Table 8. Age in days at landmark events of eye opening and primary feather eruption in 1986. Data are from the Pirlot Road test plot and Tachycineta Meadows control plot. Sample sizes are numbers of individual young, and means are compared using t-tests with pooled standard deviations (SD). Day of hatching is defined as day zero.

	EYE OPENING			PRIMARY ERUPTION		
	$\bar{X}$	SD	N	$\bar{X}$	SD	N
TEST	5.1	0.91	45	8.7	0.97	29
CONTROL	5.5	1.41	62	9.4	0.89	42
t-VALUE	1.95	NS		3.33 (P < .01)		

Table 9. Exposure data and numbers of individuals dying for eggs, nestlings, and nests in 1986 calculated using the Mayfield method (Mayfield 1961, 1975). Data are pooled from all test and control plots. Comparisons between test and control were calculated using G-tests (Sokal and Rohlf 1981).

	** EGG MORTALITY **		
	EGG EXPOSURE DAYS	EGG MORTALITIES	
TEST PLOTS	5821	139	$P < 0.001$
CONTROL PLOTS	7265	248	
	** NESTLING MORTALITY **		
	NESTLING EXPOSURE DAYS	NESTLING MORTALITIES	
TEST PLOTS	2377	135	$P < 0.001$
CONTROL PLOTS	3100	104	
	** OVERALL NEST MORTALITY **		
	NEST EXPOSURE DAYS	NEST MORTALITIES	
TEST PLOTS	1780	45	NS
CONTROL PLOTS	2385	64	
	** INCUBATION PHASE NEST MORTALITY **		
	NEST EXPOSURE DAYS	NEST MORTALITIES	
TEST PLOTS	1242	21	NS
CONTROL PLOTS	1628	39	
	** NESTLING PHASE NEST MORTALITY **		
	NEST EXPOSURE DAYS	NEST MORTALITIES	
TEST PLOTS	539	24	NS
CONTROL PLOTS	757	25	

Table 10. Comparison of R^2 values for growth rates of tree swallows during 1986.

Measure	Model[a]	Number of individuals	R2
Wing length	Exponential	59	0.9764
Tarsus length	Logistic	59	0.7929
	Von Bertalanffy	59	0.7773
Ulna length	Logistic	59	0.9255
	Von Bertalanffy	59	0.8869
Weight	Logistic	59	0.9046
	Von Bertalanffy	59	0.8521

[a] Model description: Exponential $Y = \exp(kt)$; Logistic $Y = A / \{1 + \exp[-k(t-I)]\}$; Von Bertalanffy $Y = A\{1 - [.333 \exp(-k(t - I))]\}$; where k = growth the constant, t = age in days, A = the asymptotic value of Y (here A = the maximum value for Y + 0.1) and I = the inflection point in days. k and I were determined using linear regressions of log-transformed data; transformed Y' values were ln (Y), ln [(A/Y) - 1] and ln [3(1 - $(Y/A)^{.333}$] for the exponential, logistic and Von Bertalanffy models, respectively.

Table 11a. Nested ANOVA for the effects of plot (test or control), location within a plot (LOCATION) and nest (NESTNO) on weight increase in tree swallows during 1986 (logistic model for growth constant; excluding individual growth rates which did not differ significantly from 0.0).

SOURCE	DF	SUM OF SQUARES	MEAN SQUARE	F VALUE	P > F
MODEL	13	0.13115475	0.01008883	5.36	0.0001
ERROR	45	0.08466728	0.00188150		
CORRECTED TOTAL	58	0.21582203			

R-SQUARE	ROOT MSE
0.608	0.043376

SOURCE	DF	TYPE I SS	F VALUE	P > F
PLOT	1	0.04888666	7.13	0.057
NEST(PLOT)	12	0.08226809	3.64	0.0008

Table 11b. Nested ANOVA for the effects of plot (test or control), location within a plot (LOCATION) and nest (NESTNO) on the inflection point for weight increase in tree swallows during 1986 (logistic model; excluding inflection points which did not differ significantly from 0.0).

SOURCE	DF	SUM OF SQUARES	MEAN SQUARE	F VALUE	P > F
MODEL	13	31.20973583	2.40074891	5.65	0.0001
ERROR	45	19.12784069	0.42506313		
CORRECTED TOTAL	58	50.33757652			

R-SQUARE	ROOT MSE
0.620009	0..65196865

SOURCE	DF	TYPE I SS	F VALUE	PR > F
PLOT	1	12.50140725	4.01	0.10
NESTNO(PLOT)	6	18.70832859	3.67	0.0007

Table 12a. Nested ANOVA for the effects of plot (test or control), location within a plot (LOCATION) and nest (NESTNO) on tarsus growth in tree swallows during 1986 (logistic model for growth constant; excluding individual growth rates which did not differ significantly from 0.0).

SOURCE	DF	SUM OF SQUARES	MEAN SQUARE	F VALUE	P > F
MODEL	13	0.04333887	0.00333376	5.82	0.0001
ERROR	44	0.02521362	0.00057304		
CORRECTED TOTAL	57	0.06855248			

R-SQUARE	ROOT MSE
0.632200	0.02393819

SOURCE	DF	TYPE I SS	F VALUE	PR > F
PLOT	1	0.00523391	1.65	0.20
NESTNO(PLOT)	12	0.03810495	5.54	0.0001

Table 12b. Nested ANOVA for the effects of plot (test or control), location within a plot (LOCATION) and nest (NESTNO) on the inflection point for tarsus growth in tree swallows during 1986 (logistic model). (Note small N due to exclusion of all birds with inflection point not significantly greater than 0.0).

SOURCE	DF	SUM OF SQUARES	MEAN SQUARE	F VALUE	P > F
MODEL	7	3.39096919	0.48442417	5.82	0.012
ERROR	8	0.66603227	0.08325403		
CORRECTED TOTAL	15	4.05700146			

R-SQUARE	ROOT MSE
0.835831	0.28853775

SOURCE	DF	TYPE I SS	F VALUE	PR > F
PLOT	1	0.32398618	0.63	0.50
NESTNO(PLOT)	6	3.06698301	6.14	0.0112

Table 13a. Nested ANOVA for the effects of plot (test or control), location within a plot (LOCATION) and nest (NESTNO) on ulna growth in tree swallows during 1986 (logistic model for growth constant; excluding individual growth rates which did not differ significantly from 0.0).

SOURCE	DF	SUM OF SQUARES	MEAN SQUARE	F VALUE	P > F
MODEL	13	0.06512783	0.00500983	4.08	0.0002
ERROR	45	0.05527508	0.00122834		
CORRECTED TOTAL	58	0.12040292			

R-SQUARE	ROOT MSE
0.54	0.035

SOURCE	DF	TYPE I SS	F VALUE	P > F
PLOT	1	0.00470361	0.93	0.50
NESTNO(PLOT)	12	0.06042422	4.10	0.0003

Table 13b. Nested ANOVA for the effects of plot (test or control), location within a plot (LOCATION) and nest (NESTNO) on the inflection point for ulna growth in tree swallows during 1986 (logistic model; excluding inflection points which did not differ significantly from 0.0).

SOURCE	DF	SUM OF SQUARES	MEAN SQUARE	F VALUE	P > F
MODEL	13	12.02745266	0.92518867	4.73	0.0001
ERROR	45	8.80062915	0.19556954		
CORRECTED TOTAL	58	20.82808181			

R-SQUARE	ROOT MSE
0.577463	0.44223245

SOURCE	DF	TYPE I SS	F VALUE	PR > F
PLOT	1	1.09972627	1.21	0.50
NESTNO(PLOT)	12	10.92772639	4.66	0.0001

Table 14a. Nested ANOVA for the effects of plot (test or control), location within a plot (LOCATION) and nest (NESTNO) on wing growth in tree swallows during 1986 (exponential model for growth constant; excluding individual growth rates which did not differ significantly from 0.0).

SOURCE	DF	SUM OF SQUARES	MEAN SQUARE	F VALUE	P > F
MODEL	13	0.00556851	0.00042835	17.65	0.0001
ERROR	45	0.00109233	0.00002427		
CORRECTED TOTAL	58	0.00666085			

R-SQUARE	ROOT MSE
0.836	0.0049

SOURCE	DF	TYPE I SS	F VALUE	P > F
PLOT	1	0.00007155	0.16	0.50
NESTNO(PLOT)	12	0.00549697	18.87	0.0001

Table 14b. Nested ANOVA for the effects of plot (test or control), location within a plot (LOCATION) and nest (NESTNO) on the inflection point for wing growth in tree swallows during 1986 (exponential model; excluding inflection points which did not differ significantly from 0.0).

SOURCE	DF	SUM OF SQUARES	MEAN SQUARE	F VALUE	P > F
MODEL	13	67.66387964	5.20491382	8.44	0.0001
ERROR	45	27.75208222	0.61671294		
CORRECTED TOTAL	58	95.41596187			

R-SQUARE	ROOT MSE
0.709146	0.7831073

SOURCE	DF	TYPE I SS	F VALUE	PR > F
PLOT	1	0.04907802	0.01	0.78
NESTNO(PLOT)	12	67.61480162	9.14	0.0001

Table 15. Mean values for growth constants and inflection points derived from fitted growth curves. Data are from test and control plots, 1986. See Table 10 for growth models and regression parameters.

Growth Constant:

	TEST				CONTROL			
Variable	N	$\bar{X}$	SD	CV	N	$\bar{X}$	SD	CV
WEIGHT	18	0.36	0.055	15.28	41	0.30	0.054	17.83
TARSUS	17	0.17	0.023	14.43	41	0.19	0.037	19.72
ULNA	18	0.34	0.033	9.81	41	0.32	0.049	15.48
WING	18	0.17	0.009	5.33	41	0.17	0.011	6.82

Inflection Point:

	TEST				CONTROL			
Variable	N	$\bar{X}$	SD	CV	N	$\bar{X}$	SD	CV
WEIGHT	18	7.02	0.585	8.34	41	8.02	0.895	11.15
TARSUS[a]								
ULNA	18	5.58	0.500	8.95	41	5.88	0.622	10.58
WING[b]								

[a] Inflection point included because the sample size was too low.

[b] Inflection point not applicable to curves for wing growth.

Table 16. Statistics for incubation during daylight hours for a single female on test and control plot, 1986.

	INCUBATION VARIABLES								
	% time/day			min/bout			T egg (°C)		
Nest No	Days	$\bar{X}$	SD	N	$\bar{X}$	SD	N	$\bar{X}$	SD
472 (Control)	6	81.8	10.17	63	25.95	54.27	63	34.6	2.46
274 (Test)	11	82.1	8.48	262	19.40	25.84	262	36.1	1.66

Table 17. Visits to the nest per hour for male and female tree swallows during the nestling stage (days 0 to 18 after hatching). Both nests were located at Panola Plains control plot.

Nest Number	Sex	No. of Days	Mean Visits/Hr	S.D.	Coefficient of variation
445	F	9	6.72	1.151	22.5%
	M	9	4.87	1.096	22.5%
471	F	10	5.99	1.060	17.7%
	M	10	8.30	0.925	11.1%
445 + 471	F	19	6.34	0.260	17.9%
	M	19	6.68	2.013	30.2%

Table 18a. Results of linear regression analyses of growth of deermice in enclosures at Michigamme control and Pirlot Road test during 1986.[a]

Female No	Litter Size	Start Date	Regression Statistics N	R^2	b*	(SE)
MICHIGAMME (control)						
1001	5	5/11	80	.76	.305	(.0193)
1004	6	5/11	60	.58	.258	(.0286)
1011	6	5/03	42	.94	.343	(.0142)
1012	6	6/22	47	.92	.280	(.0125)
1014	6	5/11	96	.40	.348	(.0443)
1023	4	5/22	32	.65	.302	(.0409)
1030	6	7/25	48	.56	.258	(.0335)
1042	5	7/08	43	.22	.126	(.0374)
1102	6	8/03	54	.50	.228	(.0316)
ALL FEMALES			502	.51	.284	(.0120)
PIRLOT ROAD (test)						
1010	7	5/23	49	.80	.237	(.0173)
1051	7	5/03	61	.77	.281	(.0200)
2012	7	6/06	28	.45	.421	(.0905)
2025	5	7/10	40	.20	.160	(.0524)
2041	5	7/12	45	.78	.267	(.0215)
2200	6	8/05	32	.04	.109	(.0926)
2305	5	8/01	34	.39	.220	(.0487)
ALL FEMALES			289	.54	.251	(.0140)

[a] "Female No" = identification number (toe-clip) of the mother; "Litter " = the number young in the litter for this female.

* b = the slope of the regression line and SE = the standard error of the slope.

Table 18b. Nested ANOVA of deermice growth rates on test and control plots in 1986. Mother refers of growth rates of litter mates of a particular female deermouse.

Source	df	SS	MS	F	P > F
AMONG SUBGROUPS	15	0.186961	0.012464	10.545	0.0001
PLOT	1	0.007347	0.007347	0.573	0.50
MOTHER	14	0.179614	0.012830	10.854	0.0001
ERROR	56	0.068547	0.001182		

Table 19. Relevant statistics for litter size and age of eye-opening and incisor eruption for deermice reared in enclosures and holding facility at the Pirlot Road test site during 1986.

	CONTROL				TEST			
Characteristic	N	$\bar{X}$	SD	CV	N	$\bar{X}$	SD	CV
Incisors erupt (days)	21	5.8	0.768	16.8%	20	5.5	0.826	18.5%
Eyes open (days)	28	15.8	0.669	12.7%	17	14.1	1.167	28.3%
Litter size	11	5.5	0.688	20.7%	8	5.5	1.225	50.0%

Table 20. Results of the 1986 tree swallow homing study. Data are from Panola Plains control plot and pooled from Cleveland Homestead and North Turner test plots. All times are in minutes from release. Returns are those birds which returned to the nesting area in less than 300 minutes. Likelihood to return was assessed using the chi-squared statistic and mean return times were compared with a t-test using pooled standard deviations (SD).

			RETURN TIMES		
	RETURN	NOT RETURN	$\bar{X}$	SD	N
TEST	26	3	149.8	52.6	26
CONTROL	24	7	176.9	67.0	22[a]
	X^2 = 1.6	NS	t = 1.6		NS

[a] Only 22 of 24 returns were used in this analysis because two returns had inaccurate times recorded.

Table 21. Results of the small mammal homing studies at Pirlot Road test site and Michigamme control site during the summer of 1986.

CHIPMUNKS	RETURN	NOT RETURN	TOTAL
PRT TEST PLOT	13	6	19
MGE CONTROL PLOT	20	2	22
	X^2 = 3.16	$P > 0.05$	
DEERMICE	**RETURN**	**NOT RETURN**	**TOTAL**
PRT TEST PLOT	5	1	6
MGE CONTROL PLOT	1	2	3
	X^2 = 1.82	$P > 0.1$	

Table 22. Chi-square Analysis of Tree Swallow Embryo Data from the Floodwood Site.

Year	1983 (d^2/e)	1984 (d^2/e)	1985 (d^2/e)	Total (d^2/e)
Normal	65 (0.057)	73 (0.096)	70 (0.272)	208 (0.425)
Abnormal	12 (0.260)	13 (0.433)	21 (1.227)	46 (1.920)
Total	77 (0.317)	86 (0.529)	91 (1.449)	254 (2.345)*

* Chi-square with 2 degrees of freedom = 5.991 at P = 0.05. Thus, the frequencies of developmental abnormalities are not significantly different over years for the Floodwood site. In this analysis, abnormal refers to embryos with any observable morphological abnormality. In addition, we have added the small number of infertile eggs into the abnormal class.

Table 23. Summary of embryo data from tree swallows collected over a four year period (1983 to 1986) from various sites in the immediate area of the antenna system in the Upper Peninsula of Michigan.

Location	N	% Abnormal	Chi-Square	% Infertile	Chi-Square
Pooled Sites*	188	8.0	0.00	0.00	0.00
Floodwood	254	18.1	9.25**	0.79	1.34
FNT & FST	18	34.9	16.75**	5.56***	8.42**

* Pooled sites represent the data pooled from TMC, PPC, PRT and CHT plots. The frequencies of abnormalities for these plots are homogeneous over plots and years and appear to be the basal rates. Assuming that our pooled rates are basal, we compared this pooled population with the population from Floodwood and FST & FNT using a contengency Chi-square analysis with 1 degree of freedom.

** Chi-square with 1 degree of freedom = 7.879 and 10.828 at P = 0.005 and 0.001 respectively.

*** We have compared the frequencies of egg infertility; however, the sample sizes (0/188, 2/254 and 1/18) are too small to be certain of the differences indicated. The comparisons are again between the pooled population and the population indicated using a contengency Chi-square with one degree of freedom.

Table 24. Chi-square analysis of tree swallow embryo data from control and experimental sites, 1985 and 1986.

Year	Location	Normal (d^2/e)	Abnormal (d^2/e)	Totals (d^2/e)
1985	TMC	43 (0.088)	3 (0.737)	46 (0.825)
	PRT	42 (0.020)	4 (0.165)	46 (0.185)
	CHT	10 (0.004)	1 (0.003)	11 (0.007)
1986	PPC	49 (0.146)	3 (1.207)	52 (1.353)
	PRT	29 (0.008)	4 (0.071)	33 (0.079)
	NTT	11 (1.616)	7 (13.689)**	18 (15.305)**
Totals		184 (1.882)	22 (15.872)**	206 (17.754)**

* The sites where embryos were collected are as follows: TMC = Tachycineta Meadows Control; PRT = Pirlot Road Test; CHT = Cleveland Homestead Test; PPC = Panola Plains Control; NTT = North Turner Test.

** Chi-square with 5 degrees of freedom = 12.833 and 16.750 at P=0.025 and 0.005 respectively. Thus, the embryo data is not homobeneous. Note, a single cell causes the chi-square to be significantly different (abnormal embryos from NTT. The remainder of the cells make small, non-significant contributions to the overall Chi-square.

Table 25. Chi-square analysis of tree swallow embryo data from sites other than Floodwood and NTT using data pooled over 1985 and 1986.

	Control (d^2/e)	Test (d^2/e)	Totals (d^2/e)
Normal	92 (0.036)	81 (0.039)	173 (0.075)
Abnormal	6 (0.415)	9 (0.450)	15 (0.865)
Totals	98 (0.451)	90 (0.489)	188 (0.940)*

* Chi-square with 1 degree of freedom = 3.841 at P = 0.05. Thus, the frequency of developmental abnormalities does not appear to differ when comparing control plots (TMC and PPC) and test plots (PRT and CHT). The data were pooled over two seasons (1985 and 1986) since there were no differences between seasons.

Table 26. Tree Swallow Embryos from Control (PPC) and Experimental (PRT) Plots During the 1986 Season.

Plot	Nest Number	Normal	Normal but Retarded	Abnormal	Infertile	Totals
PPC	403	4	0	0	0	4
	410	4	1	0	0	5
	418	4	1	0	0	5
	423	4	1	1	0	6
	426	4	0	1	0	5
	427	3	0	1	0	4
	445	5	1	0	0	6
	450	4	1	0	0	5
	456	5	1	0	0	6
	466	6	0	0	0	6
	Totals	43	6	3	0	52
PRT	133	4	2	0	0	6
	135	5	1	0	0	6
	137	4	1	1	0	6
	138	2	1	2	0	5
	140	5	1	0	0	6
	172	3	0	1	0	4
	Totals	23	6	4	0	33
FST & FNT	229	0	0	3	1	4
	238	2	0	2	0	4
	239	4	1	0	0	5
	244	4	0	1	0	5
	Totals	10	1	6	1	18

Table 27. Tree Swallow embryos from the 1986 season.

Sites	%Abnormalities including Retarded and Infertile	%Abnormalities with Infertiles
PPC, FST, FNT	26.2 (27/103)	13.6 (14/103)
PPC, FST	22.4 (19/ 85)	8.2 (7/ 85)

Hatching Results for 1985 and 1986

Sites	%Fail to Hatch
PRT, TMC (1985 and 1986)	12.8 (43/337)
PRT, TMC (1985), TMC (1986)*	14.6 (38/261)

* The PRT (1986) %Fail to hatch rate is significantly lower than the TMC rate and has been excluded from this calculation.

Table 28. Peak aerobic metabolic rates in deermice and chickadees captured at the Pirlot test plot (PRT) and Michigamme control plot (MGE) during the winter of 1986.

SPECIES STUDIED	MGE	PRT
DEERMICE		
Number studied	10	8
Mean peak rate of oxygen consumption (ml O_2/g*hr)	20.6	20.0
S.D. of peak rate of oxygen consumption	2.0	0.9
Mean weight (g)	17.9	18.1
S.D. of weight	2.5	2.5
CHICKADEES		
Number studied	9	8
Mean peak rate of oxygen consumption (ml O_2/g*hr)	25.8	24.0
S.D. of peak rate of oxygen consumption	1.9	1.9
Mean weight (g)	11.7	11.6
S.D. of weight	0.6	0.7

Table 29. Summary of data on peak rates of aerobic metabolism in 1985 and 1986. All data for 1985 were gathered on animals from test sites. The data listed here for 1986 are pooled across test and control sites.

VARIABLE	1985	1986	1985 + 1986
DEERMICE			
Number studied	7	18	25
Mean peak rate of oxygen consumption (ml O_2/g*hr)	20.7	20.3	20.5
S.D. of peak rate of oxygen consumption	2.4	1.6	1.8
CHICKADEES			
Number studied	10	17	27
Mean peak rate of oxygen consumption (ml O_2/g*hr)	24.2	24.9	24.7
S.D. of peak rate of oxygen consumption	1.7	2.0	1.9

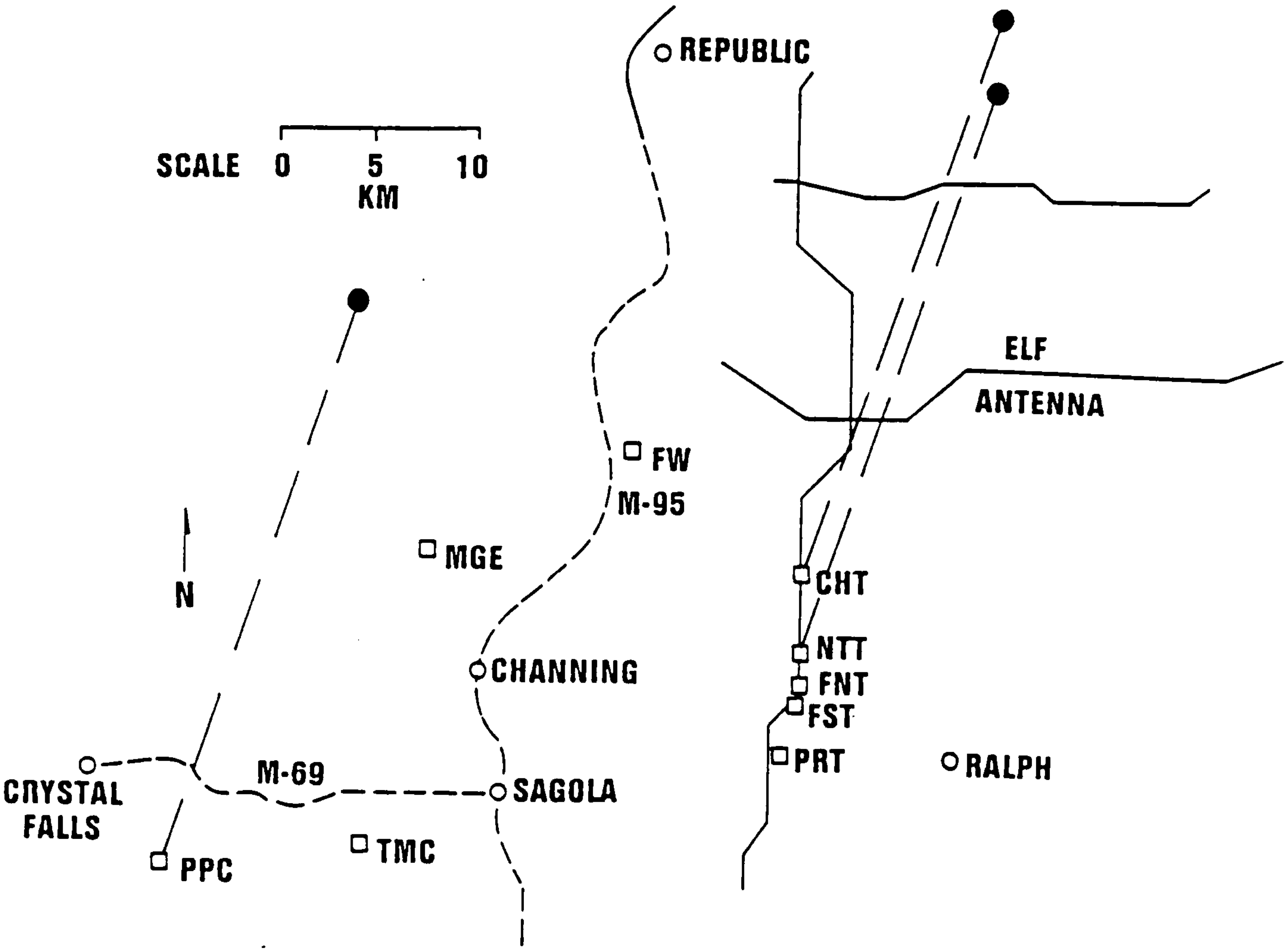

Figure 1. Test and control plots in relation to the ELF Communication System antenna line. Test plots as referred to in the text are: CHT - Cleveland Homestead; NTT - North Turner; FNT - Ford North; FST - Ford South; PRT - Pirlot Road. Control plots are : MGE - Michigamme; PPC - Panola Plains; TMC - Tachycineta Meadows. FW is Floodwood work plot which was used in the past for tree swallow studies and used in 1986 for part of the tree swallow homing study. Homing directions and release sites are also shown for tree swallows.

END

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